OmniLog® Data Collection Software
Identification System
User Guide

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Section 1. Introducing the OmniLog Identification System

The OmniLog® Identification System includes everything you need to incubate microbes and use all the features of OmniLog Data Collection software. OmniLog ID is a dedicated system for bacterial identification and includes the following components:

- Dedicated Windows®-based computer with preinstalled OmniLog Data Collection software
- OmniLog Incubator/Reader
- Printer
- Turbidimeter
- Electronic multichannel repeating pipetter

OmniLog ID system is an easy-to-use yet advanced tool for identifying and characterizing microorganisms. Our combined databases include over 1000 species of aerobic bacteria. They contain nearly all of the significant species encountered in diverse practices of microbiology, including pharmaceutical, biotechnology, cosmetic, and medical device companies; veterinary medicine; agriculture and environmental science; food processing, spoilage, and safety; reference laboratories; industrial microbiology; and research and education.

The GEN III database can continue to expand and evolve to keep pace with progress in the field of microbiology. Every month researchers discover new species of microorganisms and recognize their importance. By using its patented technology with 94 carbon source utilization or chemical sensitivity assays in a microtiter plate format (MicroPlate™), the Biolog microbial identification system can recognize over 4 x 1028 possible metabolic patterns. This provides room for future growth of the system, so that the technology will remain state-of-the-art.

How It Works

Biolog's patented technology uses each microbe's ability to use particular carbon sources or chemical sensitivity assays to produce a unique pattern or "Phenotypic Fingerprint" for that microbe. The Biolog GEN III MicroPlate provides 94 phenotypic tests: 71 carbon source utilization assays (Figure 1, columns 1-9) and 23 chemical sensitivity assays (Figure 1, columns 10-12). The test panel provides a "Phenotypic Fingerprint" of the microorganism that can then be used to identify it at the species level.

GEN III MicroPlate ™

A1 Negative Control	A2 Dextrin	A3 D-Maltose	A4 D-Trehalose	A5 D-Cellobiose	A6 Gentiobiose	A7 Sucrose	A8 D-Turanose	A9 Stachyose	A10 Positive Control	A11 pH 6	A12 pH 5
B1 D-Raffinose	B2 α-D-Lactose	B3 D-Melibiose	B4 β-Methyl-D-Glucoside	B5 D-Salicin	B6 N-Acetyl-D-Glucosamine	B7 N-Acetyl-β-D-Mannosamine	B8 N-Acetyl-D-Galactosamine	B9 N-Acetyl Neuraminic Acid	B10 1% NaCl	B11 4% NaCl	B12 8% NaCl
C1 α-D-Glucose	C2 D-Mannose	C3 D-Fructose	C4 D-Galactose	C5 3-Methyl Glucose	C6 D-Fucose	C7 L-Fucose	C8 L-Rhamnose	C9 Inosine	C10 1% Sodium Lactate	C11 Fusidic Acid	C12 D-Serine
D1 D-Sorbitol	D2 D-Mannitol	D3 D-Arabitol	D4 myo-Inositol	D5 Glycerol	D6 D-Glucose-6-PO4	D7 D-Fructose-6-PO4	D8 D-Aspartic Acid	D9 D-Serine	D10 Troleandomycin	D11 Rifamycin SV	D12 Minocycline
E1 Gelatin	E2 Glycyl-L-Proline	E3 L-Alanine	E4 L-Arginine	E5 L-Aspartic Acid	E6 L-Glutamic Acid	E7 L-Histidine	E8 L-Pyroglutamic Acid	E9 L-Serine	E10 Lincomycin	E11 Guanidine HCl	E12 Niaproof 4
F1 Pectin	F2 D-Galacturonic Acid	F3 L-Galactonic Acid Lactone	F4 D-Gluconic Acid	F5 D-Glucuronic Acid	F6 Glucuronamide	F7 Mucic Acid	F8 Quinic Acid	F9 D-Saccharic Acid	F10 Vancomycin	F11 Tetrazolium Violet	F12 Tetrazolium Blue
G1 p-Hydroxy-Phenylacetic Acid	G2 Methyl Pyruvate	G3 D-Lactic Acid Methyl Ester	G4 L-Lactic Acid	G5 Citric Acid	G6 α-Keto-Glutaric Acid	G7 D-Malic Acid	G8 L-Malic Acid	G9 Bromo-Succinic Acid	G10 Nalidixic Acid	G11 Lithium Chloride	G12 Potassium Tellurite
H1 Tween 40	H2 γ-Amino-Butryric Acid	H3 α-Hydroxy-Butyric Acid	H4 β-Hydroxy-D,L-Butyric Acid	H5 α-Keto-Butyric Acid	H6 Acetoacetic Acid	H7 Propionic Acid	H8 Acetic Acid	H9 Formic Acid	H10 Aztreonam	H11 Sodium Butyrate	H12 Sodium Bromate

Figure 1 Layout of assays in the MicroPlate.

All necessary nutrients and biochemicals are prefilled and dried into the 96 wells of the MicroPlate. Tetrazolium redox dyes are used to colorimetrically indicate utilization of the carbon sources or resistance to inhibitory chemicals.

Testing is performed simply, as shown in Section 5, Preparing Inocula. The isolate to be identified is grown on agar medium and then suspended in a special "gelling" inoculating fluid (IF) at the recommended cell density. Then the cell suspension is inoculated into the GEN III MicroPlate, 100 µl per well, and the MicroPlate is incubated to allow the phenotypic fingerprint to form.

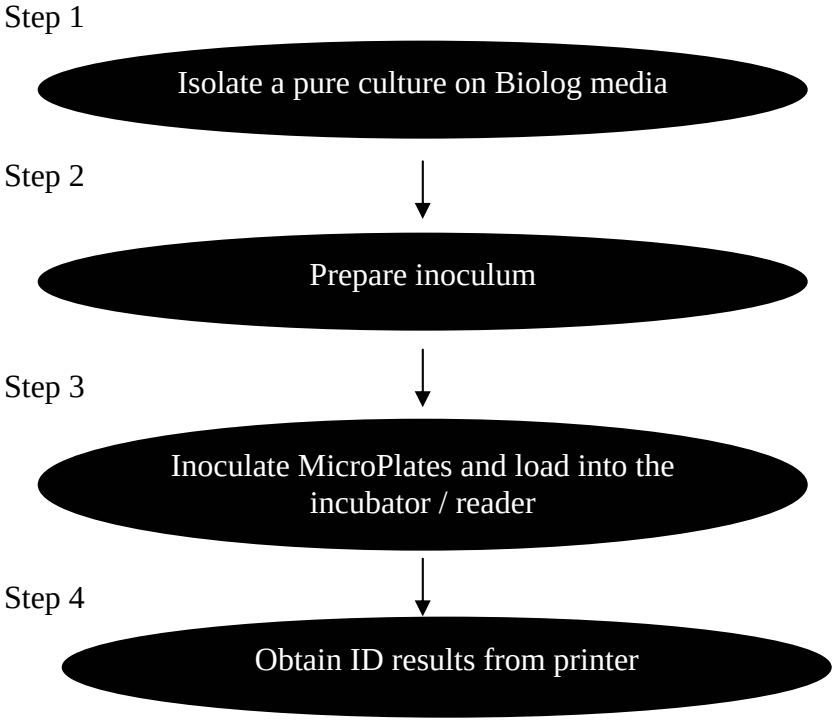
All of the wells in the MicroPlate start out colorless when inoculated. During incubation process there is increased respiration in the wells where cells can utilize a carbon source or grow in the presence of inhibitory chemicals. Increased respiration causes reduction of the tetrazolium redox dye, forming a purple color. There is a negative control well (A-1) with no carbon source used as a reference for the metabolic assays in columns 1 - 9. There is also a positive control well (A-10) used as a reference for the chemical sensitivity assays in columns 10-12.

After incubation, this phenotypic fingerprint pattern is readable by a combination incubator/reader instrument, the OmniLog Incubator/Reader. The fingerprint data is imported into OmniLog Data Collection software, which searches an extensive database and can make an identification call in seconds. By developing a simple tool that allows 94 simultaneous carbon source utilization or chemical sensitivity assays, Biolog has accomplished its goal of producing an efficient, easy-to-use, powerful, and reliable microbe identification system.

The Identification Process

Microbial identification involves four basic steps. These steps apply to all identifications. A small number of species have peculiarities that may require an extra step or special handling techniques.

The OmniLog Microbe Identification Process



Follow directions closely to obtain accurate results.

Step 1: Isolate a pure culture on Biolog media

Isolating a pure culture is not always easy. Bacteria often have sticky surfaces and cells sometimes stick together in clumps. As a first step to accurate microbe identification, streak agar plates using correct techniques to generate well isolated colonies. Always use Biolog recommended culture media and growth conditions. See Section 5 for full culturing and incubation instructions.

Step 2: Prepare inoculum

The inoculum density is very low. Gently touch the top of several colonies with the Inoculatorz™ swab. Insert the Inoculatorz into the inoculating fluid tube and emulsify the organisms into the solution using a vigorous hammering motion on the bottom of the inoculating fluid tube. This should result in an approximate % transmittance range of 90 to 98. See Section 5 for Biolog's inocula preparation directions.

Step 3: Inoculate MicroPlates and load into the OmniLog

Pipette the specified amount of cell suspension into each labeled MicroPlate, put the lid on, and place them in trays in incubator/reader. Incubate MicroPlates at 33°C.

Organisms may be incubated off line if they require high temperatures or low temperatures; in this case, you'll use the incubator/reader for reading only.

Step 4: Obtain ID results from printer

After an appropriate incubation time, OmniLog Identification System automatically reads each MicroPlate. The patterns formed in the wells are automatically entered into OmniLog Identification System, which searches the database and can provide an identification call in seconds. The printer automatically issues hardcopy results when an ID is complete.

Software Use

OmniLog Data Collection software provides an easy-to-follow visual software interface to lead the User through the identification process. There are windows for defining a Batch List, entering sample identifiers (sample number, comments, etc.), loading MicroPlates, viewing current read status, unloading MicroPlates, and viewing microbial identifications.

Before you use the OmniLog software, you must establish an ADMINISTRATOR user name and password. Please see Section 2, "Installation and Administration".

Once familiar with the system, the User will be able to prepare samples using proper techniques, read MicroPlates as a batch, and prepare batch lists to organize information.

The OmniLog Data Collection software operates the OmniLog Incubator/Reader and identifies microbes. The software is structured to move automatically through the incubation and identification process. Navigational tools include tabs to move from window to window, drop-down lists to choose from pre-set choices, selection bars, radio buttons and fields to type in specific data.

OmniLog Data Collection performs the following functions:

- Guides the User through the MicroPlate loading and reading process using Batch formats
- Guides you through unloading all or some MicroPlates when reading is complete
- Identifies microbes and allows you to interpret results

Use of Biolog's RetroSpect™ Trending and Tracking Software allows the user to manage data, edit information fields, filter data for trending and tracking analysis, compile (to develop user databases), and export the data created in the OmniLog Data Collection software.

User Functions

Logging In

OmniLog software has a predetermined time-out period. If the software is idle for more than 15 minutes, the software will Log-Out . You can prevent this logging-off by clicking "OK" when the time-out warning appears.

When the system is operating in Restricted Access mode, all users must log in when attempting to perform certain functions. The first software Log-In by a User is based on the User Name and Preliminary password given by the administrator. The software will automatically prompt them to create their own personal password. This User determined password would be used for subsequent logins.

To log in to the OmniLog Data Collection, click on the Log-In box in the upper right hand corner of the Main Screen. The Password Dialog will appear once that function is accessed.

Figure 2 Password Dialog Window

Logging Out

The OmniLog Data Collection is designed to automatically log out a user after 15 minutes of inactivity; however, to log out during a session:

- Click the Log-Out button in the upper right hand corner

Changing Users

If the OmniLog Data Collection is already open and it is necessary to change users:

1. Click Log-Out. The button will change to Log-In.
2. Click Log-In. Now, the new user will be allowed to log in using the normal log in procedure.
3. A Password Dialog window will appear. Enter the new user's name and password. Press Enter.

Changing Your Password

The OmniLog Data Collection will require users to change their password every three months, but Users may change their passwords at any time.

1. Click the Setup button on the Main screen.
2. Click the Change Password button on the bottom right corner of the Setup window under the User Options section.
3. In the Change Password Dialog window, enter your old password, new password, and new password confirmation.

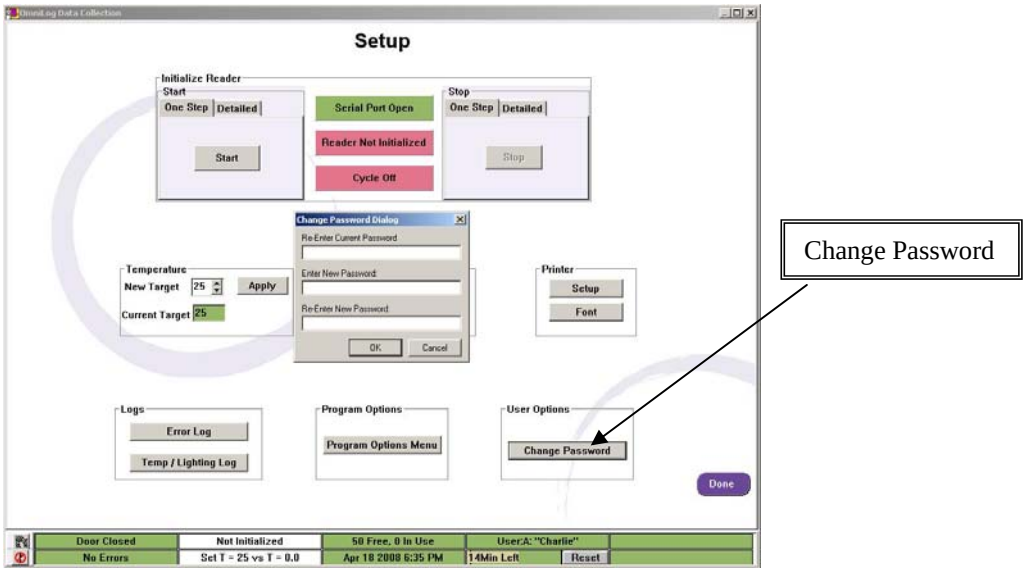


Figure 3 Setup Window with Change Password Dialog Box.

4. Click OK.

Loading and Reading MicroPlates

Batches are the backbone of OmniLog Data Collection; you'll use them throughout the process of loading and reading MicroPlates. They provide the organizational scheme for managing up to 50 MicroPlates and keeping accurate lists of identifications. Each batch can accommodate information of up to 50 MicroPlates. If you have several batches of MicroPlates in the incubator/reader at the same time, you can view results for all batches by scrolling through the list on the Read window (Figure 4). See Section 6, Loading and Reading MicroPlates for more details.

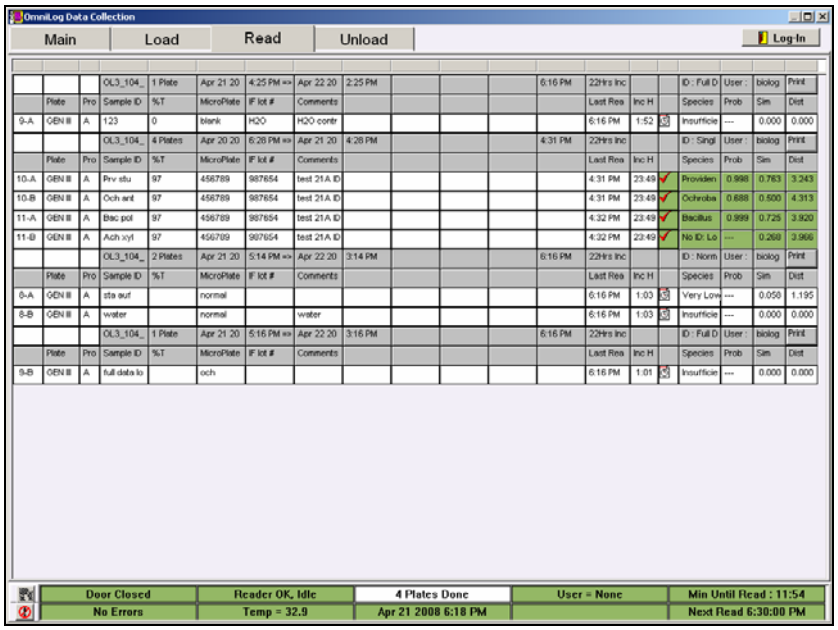


Figure 4 Read Window Showing Status of a Batch Load

In order to determine the name of your batch data file, OmniLog Data Collection directs you through entering the following information about each batch:

- Is this plate part of a project, and if so what three letters do I wish to designate as the project code?
- Do I want to use normal identification read mode or a special read mode?

As incubation and readings progress, the batches for all MicroPlates in the incubator/reader will remain displayed on the Read window. You'll be able to view in-progress and final results for all MicroPlates and all batches. When the ID process ends for each MicroPlate, you will automatically receive hardcopy results from the printer.

Interpreting Identification Results

As identification results (IDs) show up on the Read window, more detailed information can be view for any microbe listed. The Plate Data Window shows a picture of the selected MicroPlates, along with well-by-well readings, and Identification call. See Section 8, Interpreting Results for more detailed information.

Note: statistical information (optional view display) can help you assess the accuracy of the ID or pin down an uncertain ID using similarity index values and distances.

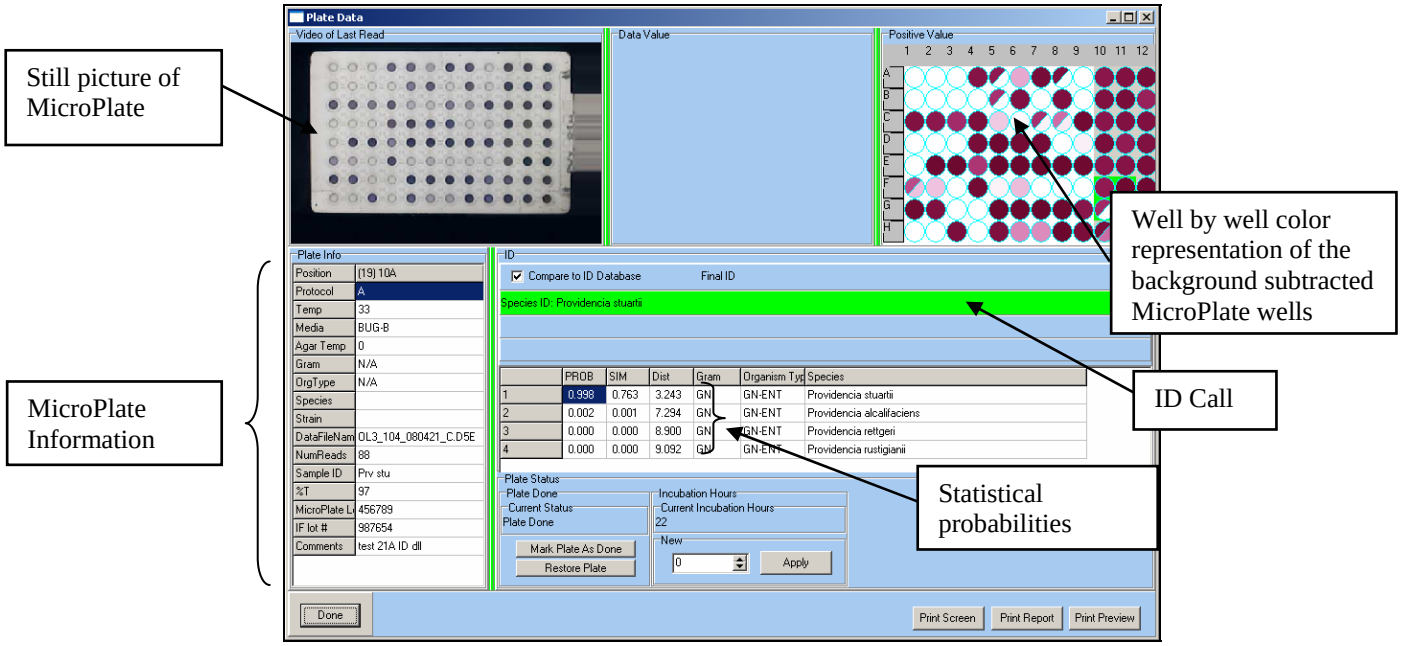


Figure 5 Plate Data Window Unloading MicroPlates

Unloading MicroPlates

After the ID process is complete for MicroPlates, they can be unload. Check the status of the MicroPlates on the Read window. Make sure all the ID final reports have printed out for those MicroPlates indicated as done. Remove the MicroPlates from the OmniLog incubator/reader, and recheck the status.

The Unload window allows you to easily tell which MicroPlates are ready to be unloaded.

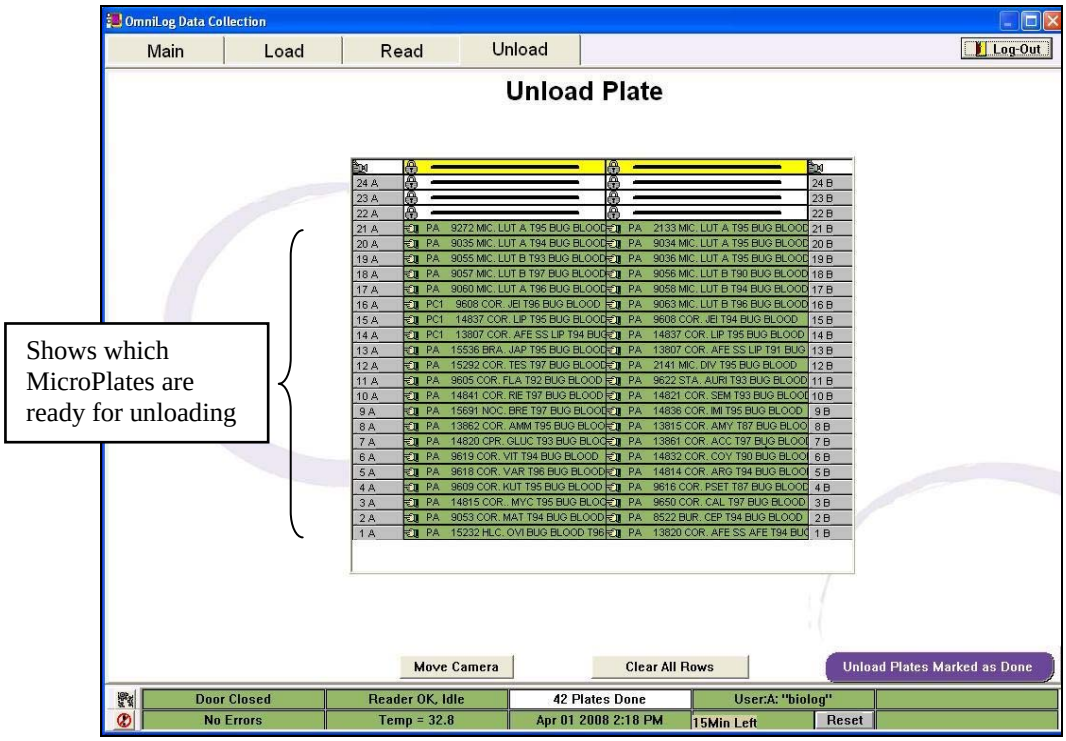


Figure 6 Unload Window

Once you ascertain which MicroPlates have completed the ID process:

- You simply open the incubator/reader door.
- Remove one tray at a time.
- Take the appropriate MicroPlate(s) out of each tray.
- Replace the tray in the same slot of the incubator/reader.
- Close the door.

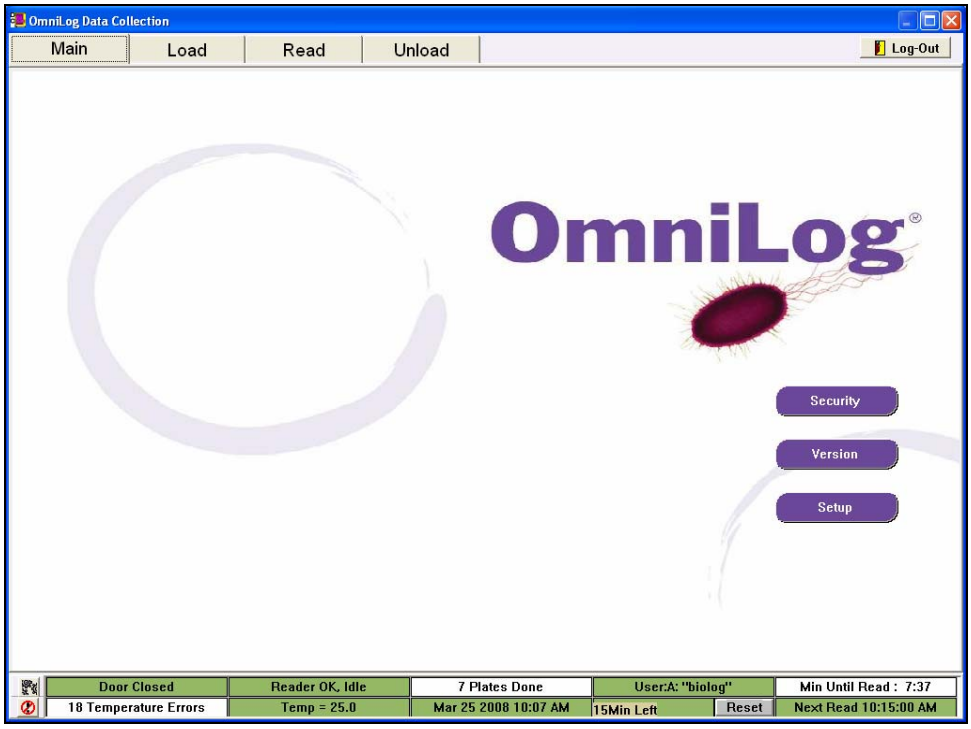
If you wish, you can load a new batch while the door is open. The system allows a minimum of 5 minutes for each load/unload cycle for every 15 minutes.

See Section 7, Unloading MicroPlates for more details.

Using the Footer Bar

No matter which OmniLog Data Collection window you display, there will always be a footer bar running across the bottom of the window. This offers a handy display of important status indicators, such as time, incubator/reader temperature, and the time until the next reading.

The footer bar consists of ten cells. Appendix 5 explains the possible entries in the footer bar cells.



Door status	Reader Status	Plate Status (Done)	User Access and Name	Minutes to Next Read
Error Messages	Temperature Status	Current System Time	Time to Auto Log Out	Time of Next Read

Figure 7 OmniLog Data Collection Footer Bar

The Math Behind the Software

OmniLog Data Collection uses extensive computer algorithms to take the information from the observed pattern and compare it to the database.

In simple terms, OmniLog Data Collection rapidly compares the positive-negative-borderline purple pattern in the MicroPlate to the species pattern in the appropriate database. The patterns that most closely match the test microbe's pattern are shown on the screen in ranked order. Before making a decision on the result, the software considers the possibility that even the first-ranked choice may not be a good match. It looks to see whether the first choice match is really "close enough" to be acceptable. If not, a "No ID" designation will result.

OmniLog Data Collection uses a newly developed pattern matching method called Progressive ID (PID). This method accurately identifies species patterns by considering the progressive sequence in which purple wells are formed. Typically microorganisms will use their favorite carbon sources most rapidly and completely, resulting in dark purple wells that form quickly. Less-preferred carbon sources are consumed slowly or incompletely, resulting in slower-forming or lighter purple color. The chemical sensitivity wells are scored positive, negative or borderline using the A10 positive control well as a reference. The extra information considered by the PID matching method brings a higher level of consistency and accuracy, representing another innovation in Biolog's technology.

Section 2. Software Installation and Configuring

Installing the OmniLog Data Collection

The OmniLog ID System is preloaded with the Data Collection software and database. Biolog's Technical Applications and Services department will help setup the system. After that, the computer and incubator/reader is generally kept on at all times for use. The installation disks that come with your system are meant for back-up only in the event of a system crash.

Note: OmniLog Data Collection works only on the computer supplied with your OmniLog ID System. Do not try to install the software on another computer.

Note:

The installation instruction is provided with the CR ROM. It is also found in the "Documents" folder on the software CD ROM or it may be requested through the Technical Applications & Services department.

General System requirements and recommendations:

Biolog recommends the use of Microsoft XP Professional, Service Pack 2 operating system.

Power settings: All power saving features must be turned off for operation of the system.

Users are required to be administrator level on Windows operating system.

The database ordered at the time of purchase will have been factory-installed. The OmniLog Data Collection software and database disc are intended for back-up only in the event of a system crash.

Software Location- The OmniLog Data Collection software must be loaded onto the computer directly attached to the OmniLog incubator/reader.

Configuring the Software

First Log-In and Setting up an Administrator

The OmniLog Data Collection is ready to be opened for the first time by the Program Administrator. The OmniLog Data Collection operates in the Restricted Access Mode. For more detailed information, see Section 4: Software Security.

The user who will act as the software administrator must implement the following steps.

1. Click on the **OmniLog** shortcut icon on your desktop.
2. The **Main** screen will appear. Click the **First Log-In** key button (Figure 8).

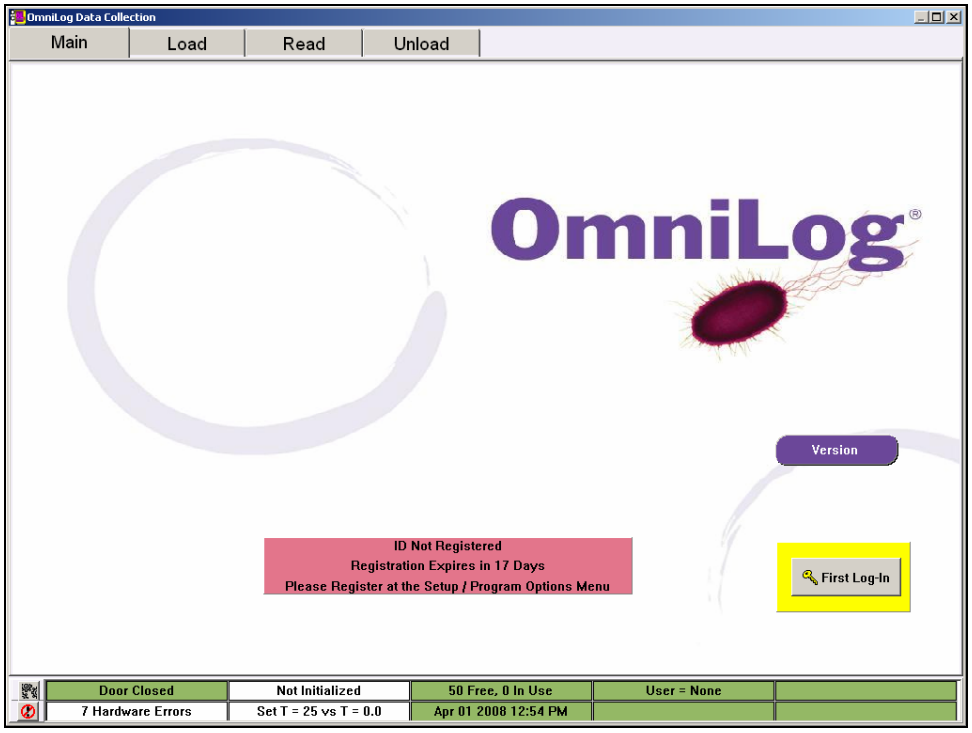


Figure 8 Main Window on first start up

3. The **Administrator Dialog** box will open (Figure 9).
   - Enter a Username that is at least 1 character in length.
   - Enter a Password that is at least 6 characters in length, alphanumeric (contains at least one number or letter). The password is case sensitive.

Figure 9 Administrator Dialog Box

4. Click **OK**. You will now be listed as the Original Administrator in the User List with full user privileges.

5. When the message to Log-In appears, click **OK**.
6. Click on the **Log-In** box located in the upper right hand corner of the **Main** window. A **Password Dialog** box will appear.
7. Enter the Administrator username and password you set in **Step 3**. Click **OK**.
8. The upper right hand of the Main screen will now show the **Log-Out** button.

Setting up the User Field List

The User Field list determines the list of fields' available to be entered for each plate record during normal operation.

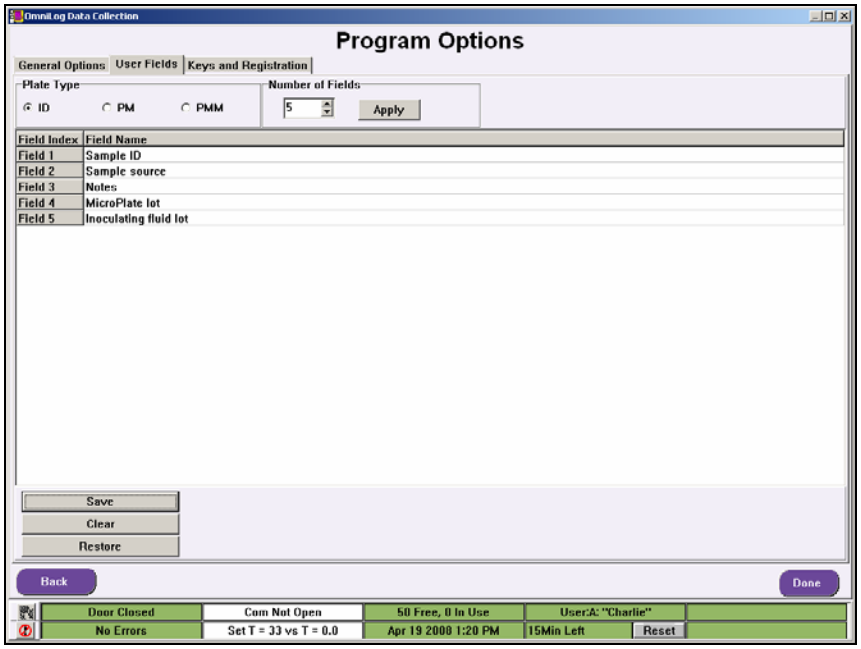


Figure 10 User Fields on Program Options Window

1. Click on the **Setup** button located on the right side of the **Main** window. The **Setup** window will appear.
2. Click the **Program Options Menu** button.

*Note: The **Program Options Menu** button can only be accessed by a user with Administrator privilege.*

3. Click on the **User Fields** tab.
4. Click the **ID** radio button if not already selected.
5. In the **Number of Fields** section, select using the arrow buttons the number of fields available to load information for each record on the **Plate Information** window.

6. Click the **Apply** button.
7. **Field 1**'s **Field Name** is currently defaulted to **Sample ID**. Depending on your needs, this can be changed.
8. Enter the **Field Name** for each of the remaining fields, as they will appear for plate information entry when creating a batch list.
    a. Examples: MicroPlate Lot; IF Lot; Media; Comment.
9. When you have completed all your **Field Name** entries you have the following options.
    a. Click the **Save** button to save your entries.
    b. Click **Clear** to clear all Field Names (to re-enter all new field names.)
    c. Click **Restore** to restore the entries in all of the Field Names to the last saved list.
10. After you have completed and saved your entries, click **Done** to accept default system configurations and display options or click on the **General Options** tab to change default settings.

System configurations: ID Database File link

This is required for the software to find the Biolog ID Database.

1. Click on the **Select ID Database File** button located in the **ID Database File** section.
2. A windows **Open** dialog box will appear. Navigate to the C:\Program Files\Biolog\OL_DB_dir\databases folder and select the database file ending with the extension .I5G (default installation location). See Figure 11.
3. Click **Open**.
4. The ID database path can be now seen above the **Select ID Database File** button.
5. If you did not find the database:
    a. Install the ID Database disc.
    b. Or browse the computer to see if the ID Database was not installed into the default location.
    c. And follow procedure as described in steps 1 thru 4 above.

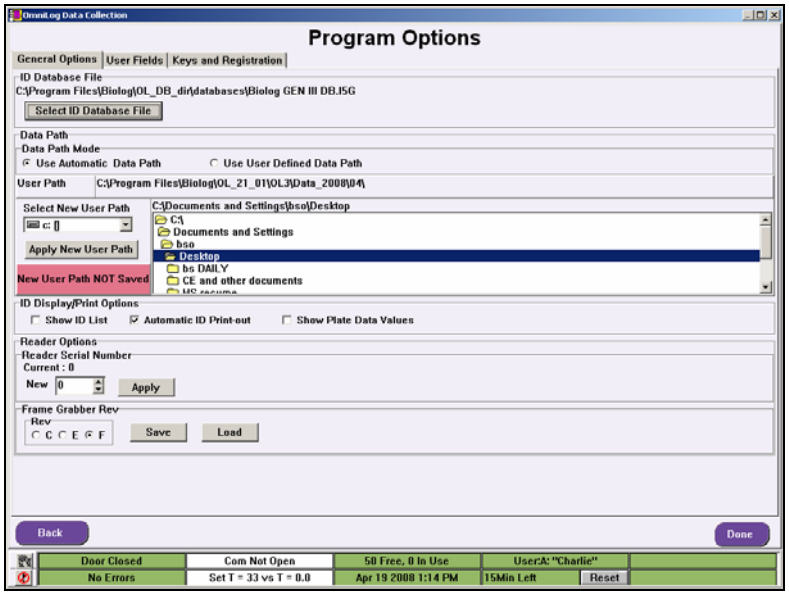


Figure 11 General Options on program Options Window

System configurations: Data Path

This section determines the location for the Data Files generated during use.

Perform this task to change the default location to a location of your choice.

Data Path Mode section:

1. Radio button selection of **Use Automatic Data Path** ignores any other settings in view and the uses the default location.
2. Current default location is C:\Program Files\Biolog\OL_XX-XX\... (XX_XX displays the current the version number of the software loaded; e.g. 21_01)
3. Radio button selection of the **User Defined Data Path** allows the user to apply settings seen to the right of the radio buttons.
4. Search and select the location for the Data Files.
5. Click **Apply New Path**.
6. The new path will display as the **Current Path**.

System configurations: ID Display and Print Options

This section determines the Display and Print options during general use.

Perform this task to change the default settings.

Show ID List selection:

1. This displays and prints in final reports the top four (4) closest matches to the ID Database and their respective ID statistics (SIM, DIST).
2. Default setting: **Not Selected**.

Automatic ID Print-out selection:

1. This sets the automatic printout of results when a final call has been made to the test MicroPlate.
2. Default setting: **Selected**.

Show Plate Data Values selection:

1. This displays a color visual 8 x 12 representation of the OmniLog reader values generated in a template form within the Plate Data pop-up window (note this does not match visual interpretation).
2. This prints the OmniLog incubator/reader values on the printout in an 8 x 12 grid.
3. Default setting: **Not Selected**.

System configurations: Reader Options

This section should already show the OmniLog reader serial number and Frame grabber rev. (video card) as applied during factory install. This section allows changes if its found to contain the incorrect information.

1. Reader Serial Number: To reset, enter or toggle to the correct number and click **Apply**.
2. Frame Grabber Rev.: To reset, select the correct Rev. letter radio button and click the **Save** button and then click the **Load** button.
3. After you have completed the **General Options** tab configurations changes, click the **Keys and Registration** tab for more configurations.

System configurations: Program Serial Number

This section allows you to correct the Program Serial Number if during installation the incorrect number was entered.

Note:

There is only 1 ID registration button access per session.

Log-Out and Log-In for additional access.

1. Click the **Enter Serial Number** button.
2. The Program Serial Number pop-up appears. Click on the blank field and enter the Program Serial Number.
3. Click **OK** to accept.
4. Click **Cancel** to go back and make a change.

System configurations: Product Key

This section should already show the acceptance of a Product Key to allow the creation of ID Batch lists that was applied during factory install. If this has not been entered contact Technical Services to obtain the Key code for your software.

System configurations: Registration

Before working with the OmniLog, it is important to register the software with Biolog Technical Services department. By registering the software, you become eligible for software upgrades and will receive regular correspondence from the company.

After initial installation, the **Main** tab will show "**Temporary Registration Days Left: 30**". The software will count down how many days you have left to register.

Registration Process

1. The Administrator will generate a **User Key** and send to Biolog
2. Load the **Registration Key** from Biolog

Generate a User Key:

1. Click the **Registration** button.
2. The **Registration Form** window appears.

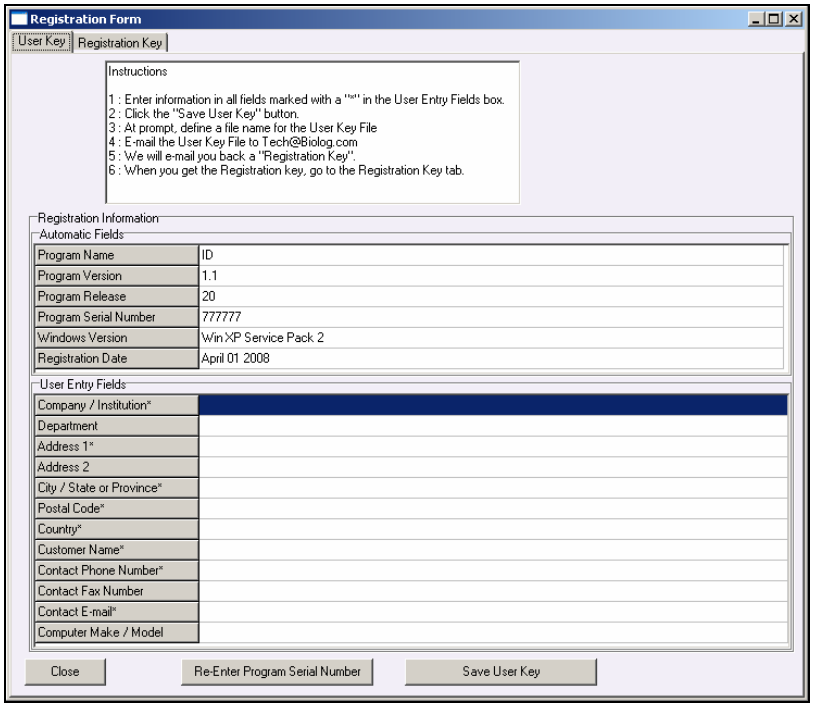


Figure 12 Registration Form

3. Fill out every entry field of the **Registration Form**. (* Required Fields)
4. Click the **Save User Key** button.
5. A **Save As** window appears. Type a file name for the User Key File in the **File Name** field. The **Save as Type** field should show Text Files as the default file type.
6. Click **Save**. The **User Key** will be saved as a text file (.txt file).
7. Click **Close**.
8. E-mail the **User Key** file to tech@biolog.com.
9. Exit and re-start the program so all of the new configuration settings are applied.

We now recommend you to read and follow **Create a User List** under the **Security** (Section 4) to input users into the system and assign privileges.

After you have e-mailed the User Key file to Technical Services and received a corresponding Registration Key, follow the instructions below to complete the registration process

Load the Registration Key:

1. Biolog will e-mail you a **Registration Key**. Save the attached registration key file on your hard drive **(...Biolog\OLDC_XX_XX\certificates).**
2. Once you have saved your **Registration key**, open the OmniLog Software, and click on the **Registration** button to open the **Registration Form**.
3. Click on the **Registration Key** tab that is on the registration form. Then click the **Load Registration Key** button to load the registration key file and process the registration.
4. Click **Close**.
5. The Temporary Registration box on the Main screen is no longer visible.

Section 3. Launching and Closing OmniLog DC

Initializing the System

In the normal course of events, the system will have been initialized during set up for continual operation. However, stopping the system maybe necessary for preventive maintenance or if you must move the OmniLog Incubator/Reader. Follow the instructions below to re-initialize the system.

1. Double-click the **OL_DC.exe** icon on your Desktop
2. The OmniLog Data Collection will start-up (Figure 13: Main window).
3. Click the **Log-In** button (upper right corner).
4. Enter **Username** and **Password**.
5. Press **Enter**.

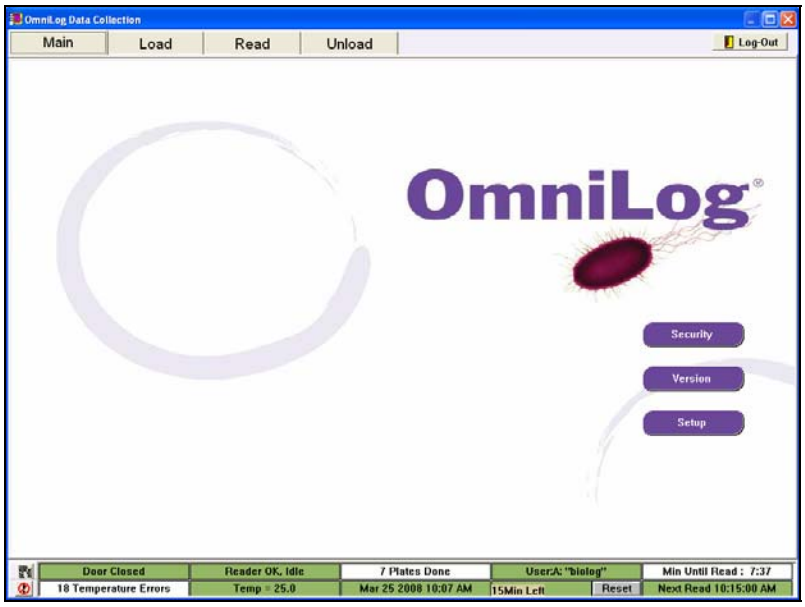


Figure 13 Main Window

The OmniLog Data Collection software will automatically connect to the OmniLog incubator/reader (initialize). Reader status indicator on the footer bar will read **Reader OK, Idle** when the process has completed.

Setting the Temperature

The Reader Setup window displays the set temperature and provides access to the temperature log of the OmniLog Incubator/Reader.

1. Set the temperature of the OmniLog to 33 degrees C, if required.
2. Click the **Setup** button on the **Main** window.
3. In the **Temperature** section of the **Setup** window (Figure 14), use the **New Target** arrows to select the target temperature.
4. Click **Apply**.
5. The **New Target** temperature will show as the **Current Target** temperature.
6. Click **Done**. The software will return to the **Main** window.

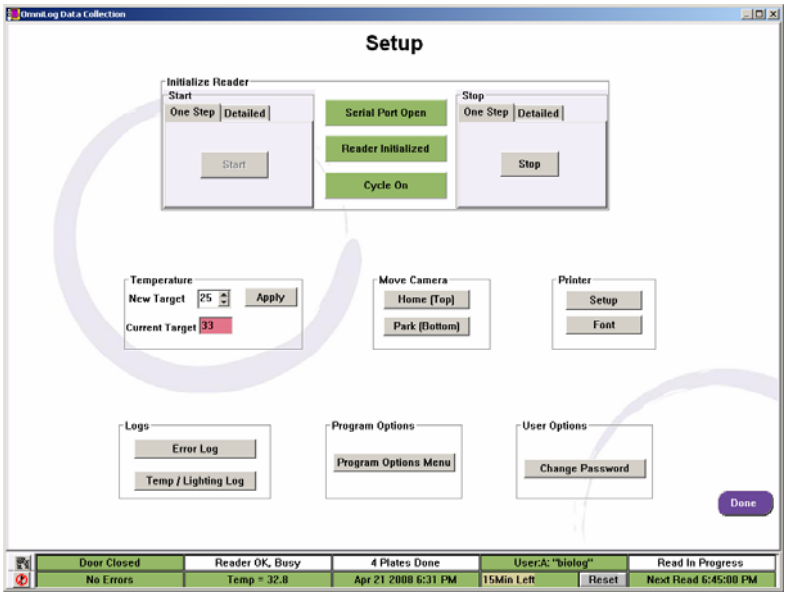


Figure 14 Setup Window

Printer settings

1. To make any changes to the printer settings:
2. Click the **Setup** button on the **Main** window.
3. In the **Setup** window, click the **Setup** button in the **Printer** section (Figure 14).
4. Make your changes and close the **Print Setup** window (Figure 15) to return to the **Setup** window.

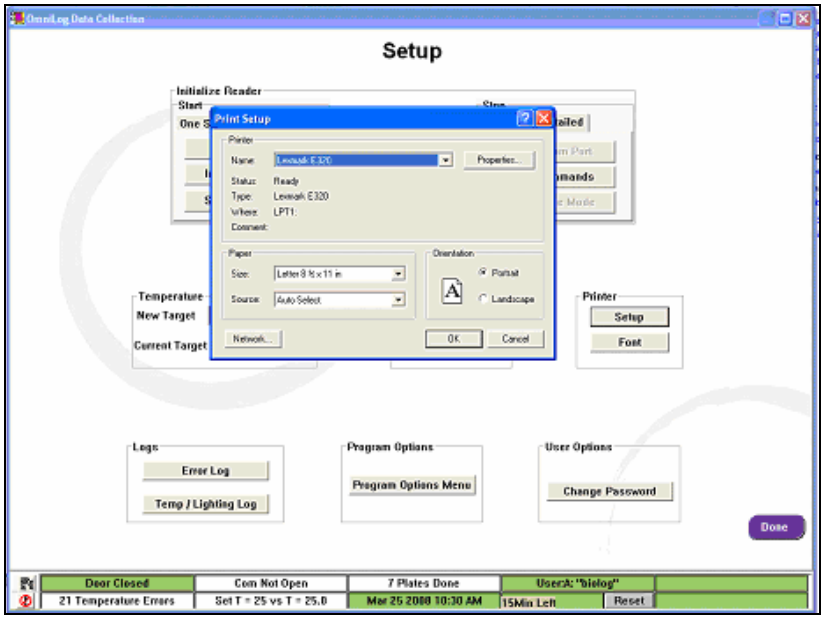


Figure 15 Printer Setup (Setup Window)

5. Click the **Font** button in the **Printer** section (Figure 14).
6. Make your changes and close the **Font** setup window(Figure 16) to return to the **Setup** window.

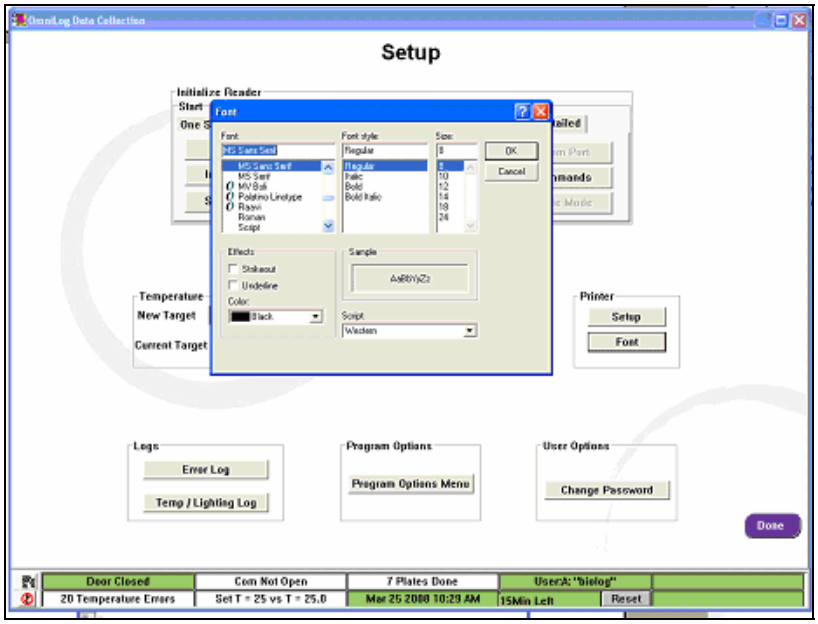


Figure 16 Font Setting (Setup Window)

Move Camera

1. To move the camera to the top or bottom tray position:
   a. Click the **Home (Top)** button to move the camera to the tray 25 position.
   b. Click the **Park (Bottom)** button to move the camera to the tray 1 position.

Shutting the System Down

Caution!

To perform an emergency stop of the OmniLog while it is moving

Click on the emergency Stop Commands button on the Status footer bar.

Exit the software by using the "**X**" close button in the upper right of the screen. This is in view with every tab screen (**Main, Load, Read, Unload**). The software will proceed to automatically close down the communication to the OmniLog incubator/reader.

1. The software will ask you to confirm that you wish to exit the program.
2. If there are plates still being processed for identification, a user must have minimally Edit privilege to close the system down and Exit with pending reads.
3. A user with minimally Edit privilege can also close down the software to OmniLog incubator/reader commands and communication as described below.
4. Under the Initialize Reader section of the Setup window:
   a. The Edit user can perform a One Step Stop.
      i. Click Stop under the One Step tab in the Stop section (Figure 17).

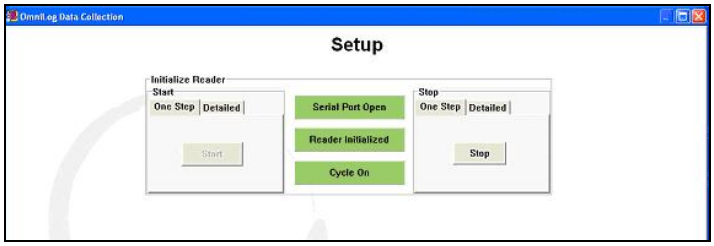


Figure 17 One Step Stop in the Setup Window

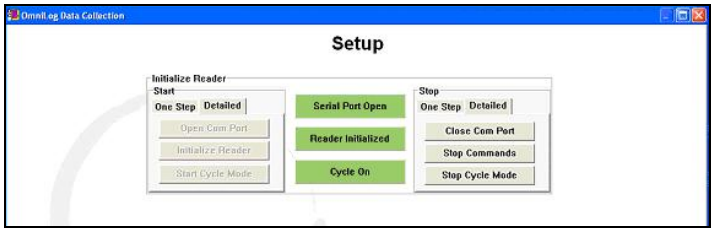


Figure 18 Detailed Stop in the Setup Window

5. The Edit user can select the Detailed tab in the Stop section (Figure 18).
    a. Click **Stop Cycle Mode**:
        i. This halts the instructions to cycle the motor to move the camera box from tray position to tray position.
    b. Click **Stop Commands**:
        i. This halts new communication between the software and the incubator/reader, but commands already in progress will continue.
    c. Click **Close Com Port**.
        i. Closing the Com (serial) port will shut down all hardware contact with the incubator/ reader.

Section 4. Software Security

The OmniLog Data Collection can be run in either Restricted Access or Unrestricted Access mode.

- Restricted Access is the recommended mode.
- Data file integrity of modified files cannot be guaranteed with Unrestricted Access mode.

What is Restricted Access Mode?

Restricted Access Mode requires that only User's with Administrative privilege oversees and controls who has access to the software. The person who is designated as the Original Administrator performed the first Log-In. This individual then manages who has access to the application and what tasks they can perform. It is recommended that more than one user be assigned Administrator Privileges on the User List.

The Administrator will:

- Assign User names and passwords for those who will use the system.
- Assign access privileges to each user.
- Oversee the security of the system

Restricted Access Mode requires all users to log in with a User ID and password when entering the OmniLog Data Collection software or changing users. The User List, Log-In Log and Log-In Log Archive files are used to keep track of registered users, access privileges, and log in/out activity in the system.

All of these files are encrypted. The Log-In Log Archive files can be placed by the Administrator in the computer/network location of their choosing. This allows the Administrator to place the files in a location that has secured access.

Why Restricted Access Mode?

Restricted Access Mode ensures that data integrity and security controls are implemented in accordance with the guidelines of 21 CFR Part 11. It assists in compliance with federal Current Good Manufacturing Practices (cGMP) by ensuring the integrity of software use and generated electronic files.

Security Features:

- User List and Log-In Log
- Notification of failed log in attempts
- User password change
- User privileges
- Session time-out
- Password expiration
- Log-In archive

Electronic record integrity:

- Limiting access
- Original record integrity
- Documentation of changes (by whom and when)
- Audit trails

OmniLog Software Flexibility in Regulated Environments

There are a number of features in the OmniLog Data Collection software that enhance utility and allow it to be easily integrated into a regulatory compliant lab. It is important that the Administrator of the OmniLog Data Collection software be aware of these features and their possible uses.

- Accessing and Managing Data- Original Data (Including Species identification) is automatically saved and cannot be manipulated in any way, by any user. However Datafiles can be used to perform a number of useful functions such as the creation of User databases and the tracking of strains (for information on these functions and other data management features please see RetroSpect Trending and Tracking Software and MicroStation Software User Guides). The chart below gives an overview of the creation and use of data.

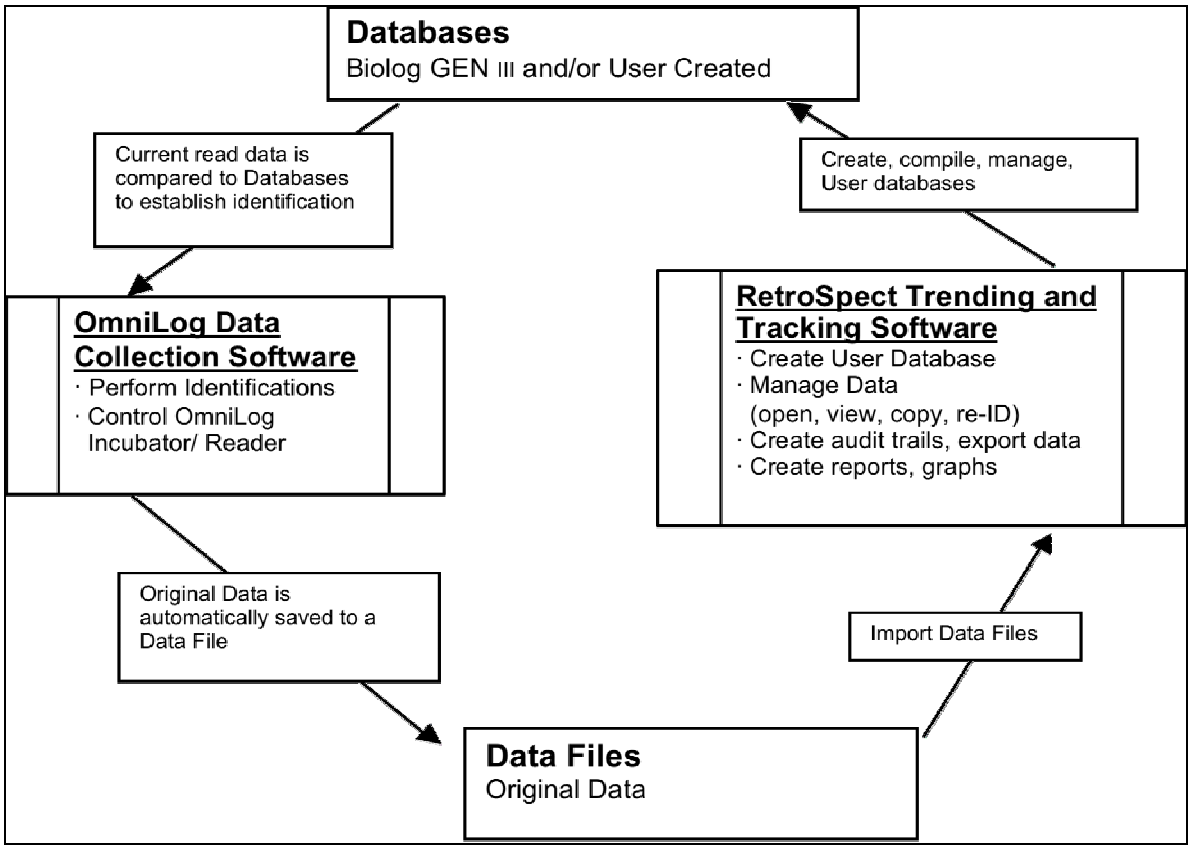


Figure 19 OmniLog Data Flowchart

What if I don't work in a 21 CFR compliant environment?

No problem! It is easy for the Administrator to assign the edit privilege to anyone in your organization who will be using the OmniLog Data Collection system. This will give everyone the ability to use all of the system's features, with the exception of the ability to access the Administrator specific functions. This allows the individual user's Username to be associated with each record to avoid confusion with multi-users in a given laboratory.

First Log-In and Setting up an Administrator

Please refer to Section 2: Installing and Configuring the Software for full instructions on how to set up an Administrator, Log In and configure the OmniLog Data Collection software for the first time. Ideally, this will be done soon after the OmniLog ID System, Data Collection has been setup and installed in your laboratory.

Administrator Functions

When a program administrator is logged in, the Administration accessible functions will be active. When other users are logged in, the Administration functions cannot be accessed.

Options Tab Functions

Once you have selected the Security button, the Administration Window will open. By default, it will appear with the Options Tab selected. Make your selections from the available options and then click the Save and Close button.

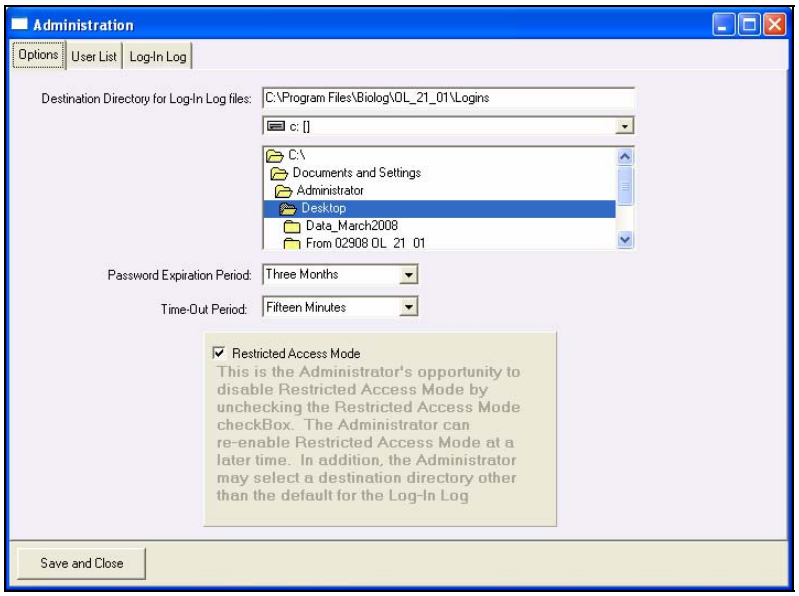


Figure 20 Options Tab in the Administration window

Important!

If you test Password Expiration Period or Time Out Period features using system clock changes, the software must be closed when system clock changes are made.

If not the Administration screen may appear with each Administrator Log-In until 100 Log-In/Log-Outs occur.

Restricted Access Mode

Default is Restricted Access Mode (check in the box). Click on the checkbox to change the mode.

- Destination Directory for LogIn-Log files
- Shows the default directory where Log-In Logs Archive files are saved. Select the desired directory to place in the computer/ network location of your choosing. You should select a secure location (for example, in a secure server or a password protected file folder).

Password Expiration Period

Use the pull down menu to select either Three Months or One Minute. The default is three months. This requires all users to select new passwords after 3 months. Select one minute to expedite validation testing only.

Time-Out Period

Use the pull down menu to select either Fifteen Minutes or 10 seconds. The default is fifteen minutes. Select 10 seconds to expedite validation testing only.

Creating a User List

Remember:

A user name must be at least 1 character in length.

The password must be at least 6 characters in length, and at least one number, and is case sensitive.

Adding new users and Assigning Privileges

1. If you are not logged in, click the Log-In button in the right upper corner of the window.
2. Enter your Username and Password (administrator) in the Password dialog box and click OK.
3. On the Main window, click the Security button. The Administration pop-up window will appear.
4. Note: Once the administrator adds a new user name to the user list, that name can never be deleted or changed.
5. Click the User List tab. A numbered list will appear, showing all users registered to that point, starting with the Original Administrator (in row 1).
6. To add a new user, click in the next blank (white) field in the User Name column.
7. Enter the new user name.
8. Click in the blank assigned Password field next to that new user name. Enter a password for that new user.

9. Click in each Privilege box to the right, toggling between Yes and No to assign or deny specific access levels to that user.
10. Click the Save and Close button when you are finished.
11. Give the User Name and Password to the person you have registered, and refer them to Logging In and Out (Section 1: starting on Pg. 6) if they need help using the OmniLog Data Collection.
12. Remember that the password the Administrator has chosen is only temporary; the new user will be prompted to enter a new password the first time they Log In to the system.

Note: We recommend that there be more than one user with administrator privilege.

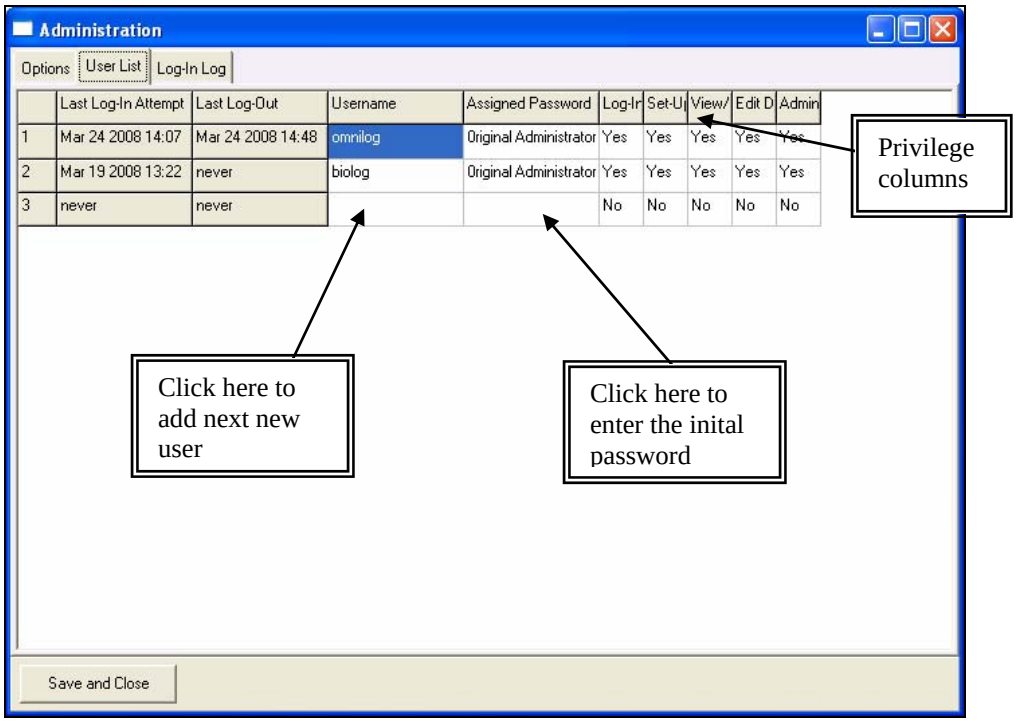


Figure 21 User List Tab in the Administration Window

Table 1 Log-In Privileges

Privilege ***Minimum User Privilege setting (Log-In, Setup, View/Print)***	What It Allows In OmniLog Data Collection
View/Print	User will be able to log onto the OmniLog Data Collection. User will be able to perform general ID functions (Reader Setup, batch, Load, Unload completed plates). User will be able to view or print data from Read Menu, view and save error logs. All three (3) privilege settings should be revoked when a user no longer uses the system.
Edit	Advance User Privilege setting (Log-In, Setup, View/Print, Edit) User will be able to Mark Plates as done, Restore plate, clear all runs in progress, Snooze Function, and change the OmniLog incubator/reader temperature or quit the OmniLog Data Collection with pending reads, clear error logs.
Admin	User will have complete access to all aspects of the software, including all Security and Administration functions.

Unauthorized Log-In Attempts

If someone enters an incorrect user name and/or password, the software will allow five attempts to enter the information correctly.

Subsequent attempts to open the OmniLog Data Collection and log in using an incorrect user name or password will result in a warning tone and screen message just above the status bar noting, "Unauthorized access has been attempted."

The message will remain until an Administrator Logs Into the OmniLog Data Collection and clicks on the warning box found after clicking the Security button within the Administration pop-up window.

If the only Administrator cannot Log-In contact Technical Services for assistance.

Lost or Revoked Password

Occasionally, a user might simply forget or misplace their password. The user may have attempted entry 5 times with the incorrect password. If the user is NOT the only Administrator, this situation is easily remedied. Any Administrator can easily go to the User List and assign the person a new password. Make sure to re-assign the user's privileges as well, or the new password will not work.

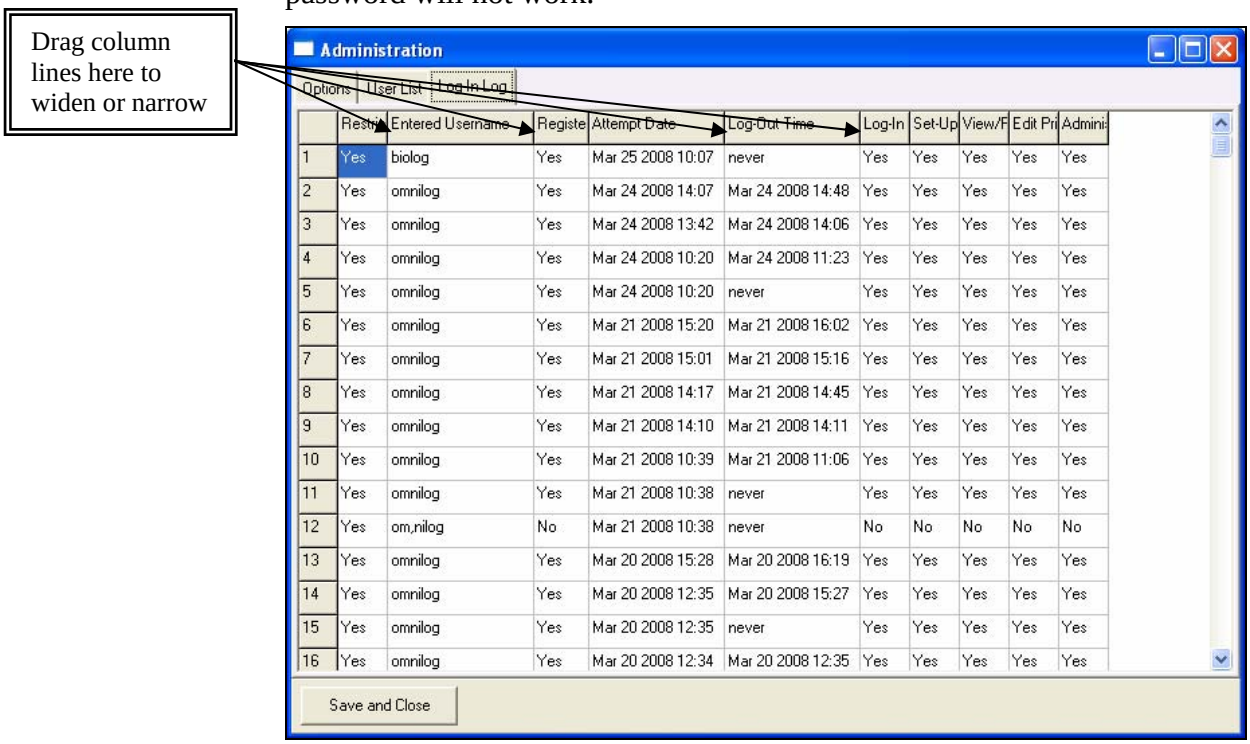


Figure 22 Log-In Log Tab in the Administration Window

Log-In Log

Only the administrator has access to the Log-In Log. This feature of the OmniLog Data Collection keeps meticulous track of the software use in descending order of date (with the most recent date and Log-In first). This log is non-editable.

- The Log-In Log records the past 100 Log-In attempts. For each Log-In attempt, the Log-In Log records the username, date and time logged-in, whether the username is registered, and the date and time the person logged-out.

- The Log also records which privileges registered users have, as well as whether the OmniLog Data Collection is in Unrestricted Access Mode. This creates an audit trail (in both Restricted and Unrestricted Access modes). As Log-In records in excess of 100 drop from the list, they are saved to a "Read-Only" file with a file name “Logins/LoginLogXXXXX.csv” where XXXXX is a five-digit date stamp. When this saved file contains 100 records, subsequent records will be saved to a new file with a different date stamp.
- Log-In-Log Storage- This location should be secure and is specified on Options Page of the Administration screens.

Table 2 Interpreting the Log-In Log

Column Name	***Information Given***
Restricted	If software was in Restricted Access mode when user logged-in, this entry will say Yes; if software was operating in Unrestricted Access mode, this entry will say No.
Entered Username	The user name of each person who logged in is listed here
Registered	If this person was an approved user, this entry will say Yes; if not, this entry will say No.
Attempt Date	This is the exact date and time the user logged in (or attempted to do so).
Log-Out Time	This is the exact date and time the user logged out. The entry here will read Never in the event of a failed Log-In and when the software administrator is currently using the software.
User Privileges	
	All User should have Log-In and Setup privileges
View/Print	Yes if access was given during that Log-In period; No to all privileges if not
Edit	Yes if access was given during that Log-In period; No if not
Admin	Yes if access was given during that Log-In period; No if not

Security Administration Suggestions

Warning:

It is important for the Administrator to use identical usernames in the OmniLog Data Collection and other Biolog software to ensure consistency of records.

The Biolog MicroStation/MicroLog and RetroSpect Trending and Tracking software include control features that ensure data integrity by limiting access, ensuring that original data cannot be changed or deleted and providing audit trails. These separate software provide greater flexibility, but they also require that the administrator control and synchronize all software. The administrator of the OmniLog software must integrate the software controls of all software into a regulatory compliant laboratory.

Usernames, Access Levels, and Passwords

The Biolog Identification software has separate User Lists. It is important to the administrator that these lists are consistent in order to create a reliable audit trail.

1. When adding a user, be sure to include a Username and privilege profile for that user in all software. Conversely, if removing a User's Access privileges, make the change in all software.
2. Use identical user names in all software for a specific user.
3. Instruct that when a user logs into the software for the first time that they Log-In and establish their own passwords in both software. By doing this you will ensure that the User's passwords will expire at the same time and eliminate possible confusion.
4. Instruct that if a User decides to change their password before the three-month expiration period, they make the change in all software.

Section 5. Preparing Samples

As with any system, the precision and accuracy of OmniLog ID results require proper sample preparation. The most common identification problems result from improper lab technique and using non-recommended media. Using good sterile technique and the correct media greatly increase the likelihood of problem-free microbe identification.

The flowchart in Sectiion 1 page 3 provides an outline of the identification process. Table 5-2 gives a comprehensive overview of the sample preparation process, as does the flowchart in Appendix 1.

Isolating a Pure Culture

Preparing an inoculum directly from a mixed-growth plate will cause identification problems. You may use a colony magnifier lamp to closely examine colonies.

1. Make certain you have a pure culture (a single organism).
2. Colonies on the plate that seem to be isolated may in fact be the result of mixed growth.
    a. This is especially true with Staphylococcus species. Careful visual examination is essential to ensuring that a culture is pure.
    b. If a colony shows any hint of pleomorphism, it is probably not a pure culture and requires additional re-streaking and isolation.
3. Carefully examine areas of confluent growth.
    a. If the lawn is not uniform in texture and color, this may indicate that the culture is not pure. Once again, re-streak for isolation.

Note: Unless your organism is a very slow grower, we don't recommend using lawn growth.

4. The opposite problem can also occur; sometimes a culture may be pure, but gives the appearance of heterogeneity.
    a. This is due to a rather common phenomenon whereby microorganisms produce more than one colony type.
    b. To be certain of its identity, purify and test each colony type individually.

Characterizing Aerobic Bacteria

Recognizing fastidious gram negatives

1. Fastidious gram-negative bacteria are primarily isolated from the respiratory tracts of mammals.
2. These bacteria grow poorly or not at all on BUG + B medium. They grow much better on chocolate agar at 35-37(C in an atmosphere of 6.5% CO2.

Note: All microbes requiring a CO2 atmosphere must be grown in a secondary or offline incubator before preparation of the inoculum.

Recognizing microaerophilic gram positives

1. Microaerophilic gram-positive bacteria are primarily isolated from mammals.
2. These bacteria grow poorly or not at all on BUG + B medium without an atmosphere of 6.5% CO2. They grow much better in an atmosphere of 6.5% CO2.

Culturing Your Microbe

Isolate a pure culture on Biolog recommended agar media (BUG+B or Chocolate Agar).

1. Most bacteria should be grown at 33(C.
    a. Some species may require special culture conditions, for example some genera may require grown at 35-37(C in elevated CO2 (6.5 to 10%).
    b. Others require growth on Chocolate agar also. A few may require 26(C incubation for growth.
2. Culture your sample on Biolog's general-purpose culture medium using a single, isolated colony.
    a. Nearly all the species in the OmniLog ID database will grow on Biolog Universal Growth media (BUG), generally with the addition of 5% sheep blood.
    b. If you have not purchased prepared media from Biolog, prepare media according to the package insert. See Section 11 for media preparation instructions.
3. Most of the species in Biolog's database are relatively fast growers.
    a. However, if your microbe grows slowly, you may need to streak (subculture) more than one agar plate.

4. Do not allow cultures to grow for too long.
    a. Maximum growth is 24 hours for most bacteria. Some exceptionally slow-growing or fastidious bacteria may require 48 hours growth, or the use of multiple growth plates.
5. Use of alternative media should be validated.
    a. For laboratories that need to use agar media without blood, we recommend using BUG Agar. However, some species will grow extremely slowly or not at all if blood is omitted and therefore will not be identified, for example the genera listed for Protocols C1 and C2 in Table 1.
    b. R2A Agar and Tryptic Soy Agar without or with blood (TSA, TSA+B) will not culture as wide a range of bacteria as BUG+B. Furthermore, their recipes and performance characteristics from different vendors may vary.

Table 3 and Appendix 1 will help you select the optimal culture medium.

Table 3 Selecting the Correct Culture Medium

Organism Type	Atmosphere/ Temperature	Culture Media
Aerobic Microorganisms	Aerobic/ 33°C	BUG + B
Gram Positive Microaerophilic	6.5% CO2/ 35-37°C	BUG + B
Gram Negative Fastidious	6.5% CO2/ 35-37°C	Chocolate

Preparing Inocula

Remember, your microbes are alive. Treat them with care. Use fresh cultures.

1. Before starting, prewarm MicroPlates and Inoculating Fluids to room temperature and review the entire protocol, including precautions.
2. Once your microbe is isolated and cultured, prepare a liquid inoculum.
3. Determine Appropriate Protocol to Use (Inoculating Fluid and Cell Density)

4. All protocols are performed in the same manner, the only difference being the choice of inoculating fluid (IF) and cell density for inoculation.
    a. Protocol A is used for the vast majority of species. The inoculum must be within the range specified. If unsure of the appropriate test protocol, use protocol A.
        i. If the result fails to yield an identification call because of a false-positive A-1 well, then use Protocol B.
        ii. If the result fails because of less-than-four positive carbon source reactions, then try, in succession, Protocols C1 and C2.
    b. Protocol B is used for a small number of strongly reducing species (primarily some Aeromonas, Vibrio, Bacillus, and Tsukamurella species).

Table 4 Selecting the Correct Inoculation Fluid and Cell Density

Organism Type	Protocol	Inoculating Fluid	Inoculum Density	Species
Aerobic Microorganisms	A	IF -A	90-98% T	Nearly all – this is the default protocol
Over reactive Micoorganisms	B	IF -B	90-98% T	Strongly reducing GN (e.g., some *Aeromonas, Vibrio*) and GP (e.g., some *Bacillus, Tsukamurella*)
Gram Positive Microaerophilic	C1	IF -C	90-98% T	Microaerophilic, capnophilic GP (e.g., *Aerococcus, Arcanobacterium, Dolosicoccus, Dolosigranulum, Eremococcus, Erysipelothrix, Gemella, Globicatella, Helcococcus, Lactobacillus, Lactococcus, Leuconostoc, Oenococcus, Pediococcus, Streptococcus, Weissella, and some Corynebacterium sp.*)
Gram Negative Fastidious	C2	IF -C	62-68% T	Fastidious, capnophilic GN (e.g., *Actinobacillus, Aggregibacter, Alysiella, Avibacterium, Capnocytophaga, Cardiobacterium, CDC Group DF-3, CDC Group EF-4, Dysgonomonas, Eikenella, Francisella, Gallibacterium, Gardnerella, Haemophilus, Histophilus, Kingella, Moraxella, Neisseria, Oligella, Simonsiella, Suttonella, and Taylorella*) and GP (*Actinomyces, Alloiococcus, Granulicatella, Lactobacillus*)

i. These strongly reducing species will give a false-positive result in the A-1 well with Protocol A. If this occurs, simply repeat the test using Protocol B.

c. Protocol C1 is used for slow growing bacteria that form pinpoint-sized colonies (less than 1 mm in diameter) on BUG+B Agar in 24 h of growth (see example in Figure 2.d.).

i. These are primarily microaerophilic and capnophilic Gram-positive cocci and tiny rods (e.g., Streptococcus, Aerococcus, Pediococcus, Lactococcus, Lactobacillus, etc.). See Table 5.2. below.

d. Protocol C2 is used for fastidious, capnophilic, and very oxygen-sensitive bacteria that grow very slowly or not at all on BUG+B Agar.

i. For example, it is used for fastidious Gram-negative species grown on Chocolate Agar with 6.5% CO2. Some very oxygen-sensitive Gram-positive bacteria also require the higher inoculation density of Protocol C2 (e.g., Actinomyces, Alloiococcus, Granulicatella, and some Lactobacillus). See Table 5.2.

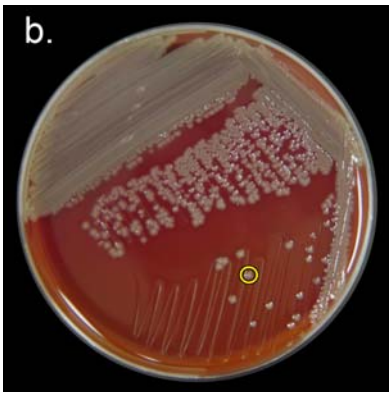
If unsure of the appropriate test protocol, use protocol A. If the result fails to yield an identification call because of a false-positive A-1 well, then use Protocol B. If the result fails because of less-than-four positive carbon source reactions, then try, in succession, Protocols C1 and C2.

Preparing inocula

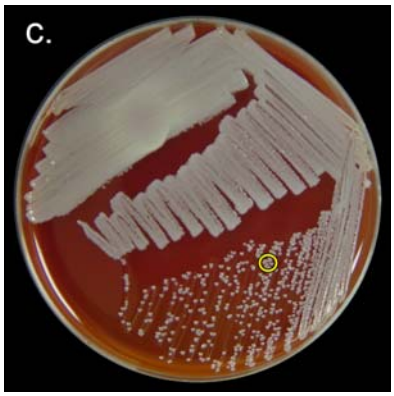
Use a calibrated Turbidimeter. Check the calibration using Biolog turbidity standards between 85 and 65% Transmittance.

1. For each inocula preparation, blank the turbidimeter with the uninoculated inoculating fluid tube (wiped clean of dirt and fingerprints) by adjusting the 100% transmittance adjustment knob so that the meter reads 100%T.
2. The inoculum density is very low. Prepare the inoculum at the desired turbidity. The target cell density should be in the range of 90-98%T for Protocols A, B, and C1. Protocol C2 requires a higher cell density of 62-68%T.
3. Use the cotton-tipped Inoculatorz to gently touch the top of a colony or colonies with the inoculatorz swab to pick up a 3 mm diameter area of cell growth from the surface of the agar plate.

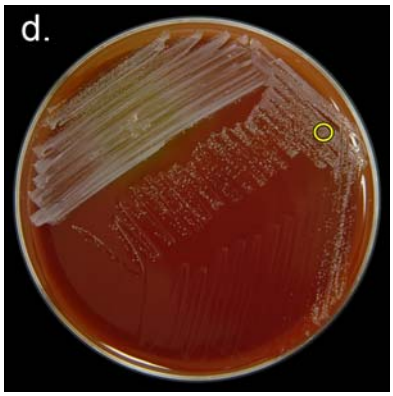
4. Grasp the swab shaft at the end, as shown (a.), and holding the swab vertically; touch it to the cell growth.
5. The following examples (b, c, d.) show photos of fast, medium, and slow growing bacteria. The yellow circle indicates where to touch the end of the cotton swab.



(b.) Fast grower



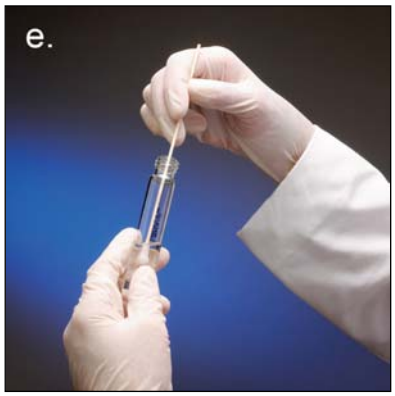
(c.) Medium grower



d.) Slow grower

6. For fast growing bacteria, large colonies: touch a single colony.
7. For medium growing bacteria, moderate colonies: touch a cluster of colonies (several colonies).
8. For slow growing bacteria, small colonies: touch the first area of confluent growth.

9. As shown below (e.); insert the Inocululatorz into the inoculating fluid tube and emulsify the organisms into the solution using a vigorous hammering motion on the bottom of the inoculating fluid tube to release the bacteria into the inoculating fluid.



10. Allow any bubble that may have formed to disperse. Read the turbidity in the turbidimeter, as shown below (f.).
11. This should result in an approximate % transmittance range of 90 to 98 for Protocols A, B, and C1 or 62-68%T for Protocol C2.
12. Adjust the inoculum density if necessary. You can change the density by adding more cells (to increase density) or more inoculating fluid (to lower density).
13. Examine your suspension and make certain that it is homogeneous and free of large clumps. If there are only a few clumps, allow them to settle to the bottom, pouring off the supernatant.

Note: For techniques to emulsify clumpy bacteria see Section 10: Technical Notes.

Inoculating MicroPlates

Note:

Pipette the inoculum into a MicroPlate within 20-30 minutes. Use sterile technique.

Inoculate the suspension into the GEN III MicroPlates.

When running a batch of MicroPlates, set them up (from preparing the inoculum to pipetting into the MicroPlate) so you will not exceed 20- 30 minutes.

Inoculating protocol

1. Label the MicroPlate with the organism name/number. Label the side of the MicroPlate itself, not the lid (Biolog imprint side).
2. Using aseptic technique, pour the cell suspension into a multichannel pipette reservoir.
3. Firmly attach eight sterile tips to the 8-Channel repeating pipetter. Refer to the pipetter manual for detailed instructions.
4. Fill the tips with the suspension. Check to see that all tips are filling equally and the tips are not leaking.
5. If you are using a manual pipetter, prime the tips by dispensing once back into the reservoir. If you are using an electronic pipetter, it will prime itself.
6. As shown below (g.), fill all MicroPlate wells with 100(L/well. Take care not to splash from one well to another. Avoid contamination.

7. Avoid touching the bottom of the wells, which could transfer carbon sources.
8. If the fluid level in the tips gets low, refill and continue dispensing until all wells are full. The electronic pipetter will double beep, signaling you to purge and refill if necessary.
9. Cover the MicroPlate with its lid.
10. The inoculating fluid will form a soft gel shortly after inoculation.

Incubating MicroPlates

As soon as you dispense the suspension, incubate the MicroPlate at 33°C. Remember, the OmniLog can only be set to one temperature per cycle. MicroPlates requiring other incubation temperatures must be incubated offline.

1. Use the OmniLog Data Collection software (as described in Section 6) to load the GEN III MicroPlate into the OmniLog as shown in picture (h.).
2. Or incubate the GEN III MicroPlates in an offline incubator.
3. You may use an off line incubator to incubate at extreme high or low temperatures or if the OmniLog is completely fully loaded. As listed in Table 5.

Table 5 Selecting the Correct Incubation Condition

Organism Type	Temperature	Atmosphere	Incubation Time
General Organisms	33° C	Air	Up to 22 hours
Organisms requiring lower temperature.	26° C	Air	Up to 22 hours Incubate in secondary incubator
Organisms requiring higher temperature.	55° C	Air	Up to 22 hours Incubate in secondary incubator
Note: Slower growers may take up to 36 hours.			

Section 6. Loading and Reading MicroPlates

Once your MicroPlates are properly inoculated (see Section 5), the next step is to enter information into Biolog's OmniLog Data Collection software. This information relates to organizing plate data in batchs, managing files, and printing results. Then you can load MicroPlates into the OmniLog incubator/reader.

OmniLog Data Collection software is an automated system that relies on proper record-keeping at the start of each batch. Be sure to proceed through these data-entry tasks with care.

The incubator/reader can hold up to 50 MicroPlates (with two MicroPlates per tray). You can load up to 50 batches of MicroPlates.

**Note: Filling out a manual batch in preparation of the setup helps with workflow. See Appendix 2 for a sample batch.*

Checking Load and Batch Status

In the normal course of operation, the Read window will be displayed when you begin the process of loading MicroPlates. The Load window shows a picture of the tray stack and indicates the current status of all MicroPlates in the incubator/reader. The Read window gives details about each MicroPlate currently in the incubator/reader.

Table 6 explains the entries you will see on this screen.

Table 6 Load Window Entries

Key	Explanation
Numbers along left and right edges	Correlate to tray numbers, starting with number 1 at the bottom through number 25 at the top
A and B designations along left and right edges	A = left column of MicroPlates B = right column of MicroPlates
White slots with ----------	Slot empty of both MicroPlates and tray
White slots with [_____]	Slot contains tray, but not MicroPlate
White background with clock icon	Slots containing plates that have not been read yet
Green background with red checkmark icon	Slots that contain MicroPlates that have been read and are ready to be removed
Yellow background with hand writing icon	Slots containing MicroPlates that have been read but are not ready to be removed. Data still saving.

Caution!

Be sure to load MicroPlates into the incubator/reader within 20-30 minutes after inoculating them. Allowing MicroPlates to sit around can cause you to miss readings.

1. Click Read on the top menu bar.
2. The Read window will appear, showing details for each MicroPlate currently in the incubator/reader.
3. Check batch status by tracking the icons to the right of the Unload column.

Figure 23 Read Window explains the columns on this screen.

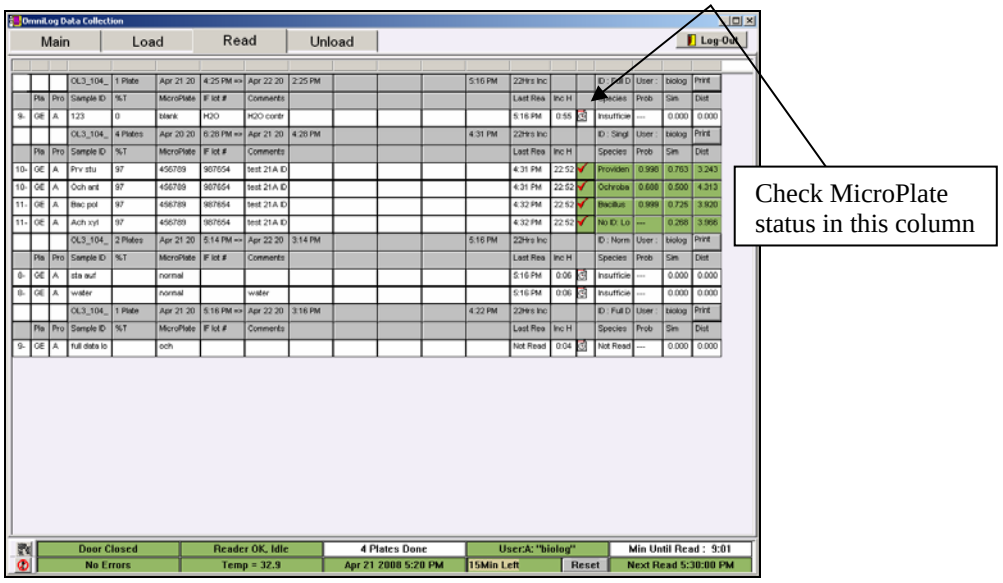


Figure 23 Read Window

Table 7 Read Window Data

Column	Explanation
Position Number	Shows batch slots as they are in the incubator/reader
Plate Type	Shows Plate type entered
Protocol	Shows Protocol entered
Sample Number	Shows sample number entered (Default User Field setting)
User field(s)	Shows User field(s)entered
Last Read	Shows the elapsed since data was taken
Incubation Hours	Shows how long the MicroPlate has been incubating
Plate Status	Shows those slots that are done and continuing reading

Setting Up a Batch List

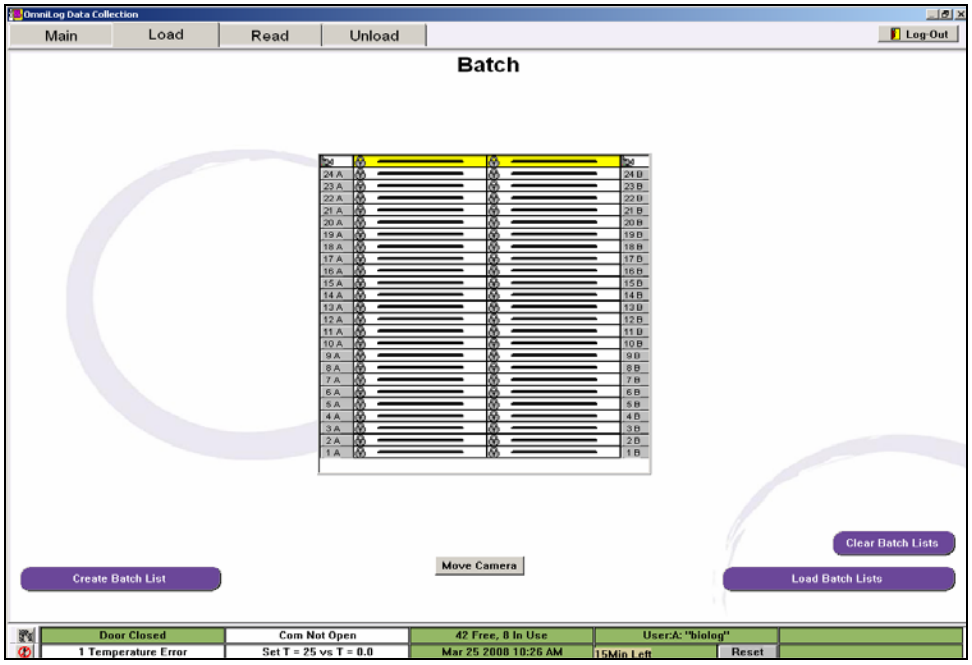


Figure 24 Batch Window

1. Click the Load tab.
2. The Batch window appears. This window allows you to create a Batch List for the MicroPlates you are about to load.
3. Click the Create Batch List button.

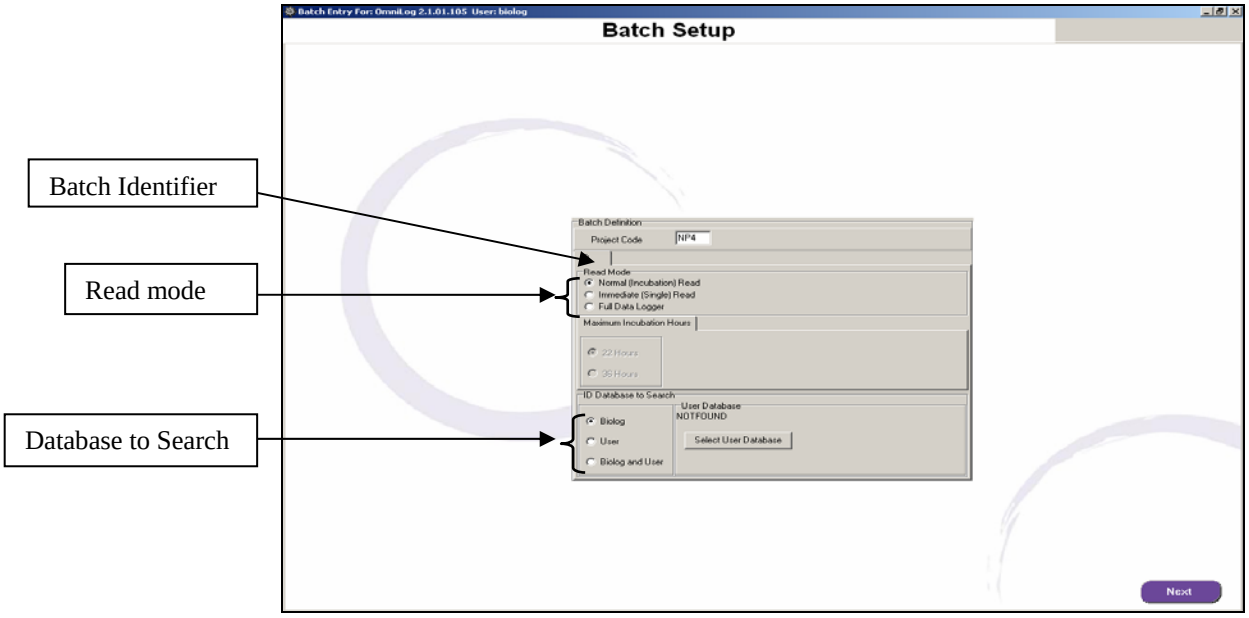


Figure 25 Batch Setup Window

4. The Batch Setup window is displayed.
5. Using the radio buttons, select the Read Mode. (Refer to Table 8 for definitions of the Read Modes.)
6. Enter a 3 character Project Code.
7. The purpose of the Project code is to give the user the opportunity of organizing data under a specific project name or number.
8. If the user does not specify a Project code, the default code of IDS will automatically be selected.

Choosing a Database to Search

1. OmniLog software allows you to select which database you want to search.
2. For information on creating a User database, see RetroSpect Trending and Tracking Software.
3. ID Database to Search selection: Select Biolog, User or Biolog and User using the radio buttons and Select User Database button, if required.
4. Click the Next button.

Note: When you're conducting a OmniLog/User database search, a "U" will appear on the Species ID ranked list on the Data window to denote species in the User database. Also, you will see a ML/user designation in the upper left corner of the organism choices.

Read Modes

Table 8 Software Read Modes

Mode	What It Does
Normal	MicroPlates are scheduled to read every 15 minutes occurring every quarter-clock hour until and identification call is made. With the recommended maximum incubation time of 22 hours, the scheduled readings stop when an ID is made, a file is saved to disk and the ID Report is printed. If no ID is found, the last read (22 hour) is saved and the ID Report is printed. When an identification is made, the MicroPlate is marked on the computer screen as finished. The MicroPlate can be removed freeing up the position in the incubator/reader.

Immediate (Single Read)	This mode is for Identification MicroPlates that are incubated off-line. All the MicroPlates in the batch will be read once immediately after loading is finished. The system will not wait for the next half clock hour. This single reading will be automatically saved to the computer disk and the ID Report is printed out. The MicroPlates are marked as finished and can be removed. When using this mode, be sure to use the correct incubation time.
Full Data Logger	This mode is useful for building a full progressive user database. MicroPlates are read every 15 minutes occurring every quarter-click hour until 22 or 36 hours as selected by the user.

Load Plate Information

Figure 26 Batch List Window

1. The **Plate Information** Window is displayed.

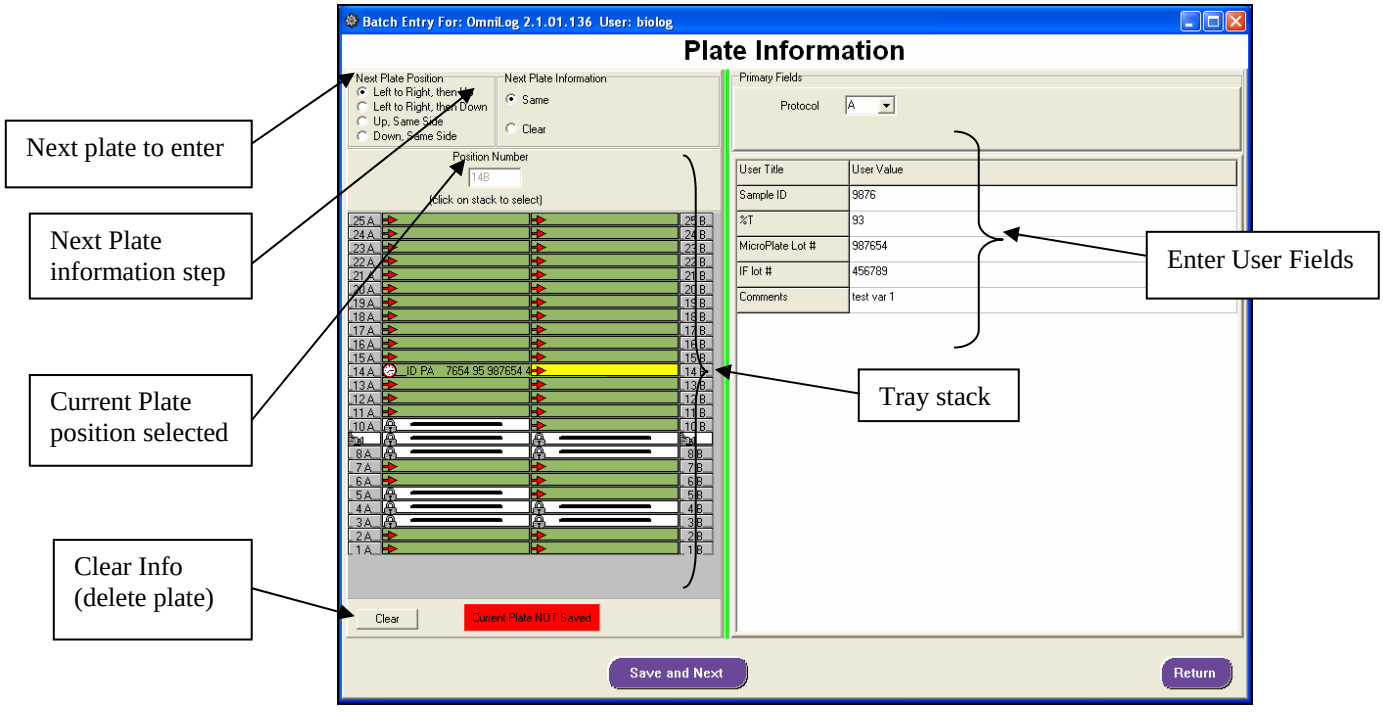


Figure 27 Plate Information Window

2. To input Plate Information after selecting the Edit Entry button, make the following flow selections:

a. Next Plate Position radio button selection determines how you want to organize the loading of your Batch List

   i. Left to Right, then Up

   ii. Left to Right, then Down

   iii. Up, Same Side

   iv. Down, Same Side

b. Next Plate Information radio button selection determines if you want to have the entire inputted field entries of the last plate remain or cleared for entry of the next plate position.

   i. Same - Used if there are many inputted field information common to each entry, user can change individual fields as needed.

   ii. Clear - Used if you want each plate information cleared before entering the next plate information.

Edit a batch while working on it by clicking on the position number of the entry you wish to edit.

3. Click the plate stack on the tray position for your first plate entry.
4. One by one, click on the blank field and enter your plate information for each field name displayed.
5. After completing the entries for the plate, make one of the following selections:

   a. Click the Save and Next button to save the plate information and enter the next tray position plate information.

   b. Or click Clear to delete the field information and tray position batch list entry.

   c. Or click Return to review entries of the Batch List.

6. After completing all entries for all MicroPlates in the Batch List, click Return and your current batch list will display for you to review and/ or print.

Note:

Be sure to print your batch and verify that all field information is correct.

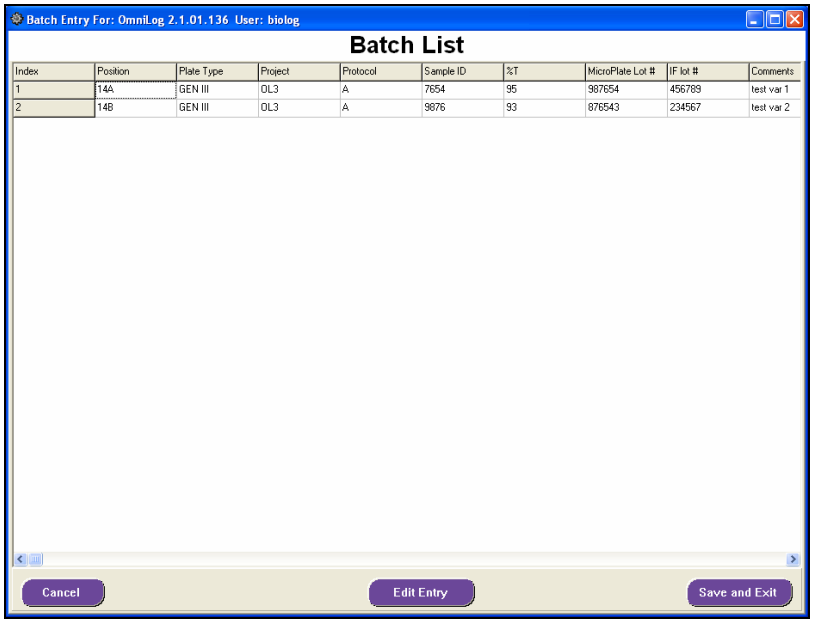


Figure 28 Batch List (with entries)

7. If there are corrections that need to be made, simply click on the Add Entry button.
   a. Click on the plate position in the stack where you want to make changes.
   b. Click on the field(s) and make your changes.
   c. Click the Save and Next button.
   d. Click the Return button when complete.
8. Your updated batch list will display for you to review and/ or print.
9. After you are assured that your Batch List entries are complete, click the Save and Exit button.
10. The Batch window is displayed.

Loading MicroPlates into the Incubator/Reader

1. At this point you may do one of the following:
   a. Create another Batch List:
      i. Click the Create Batch List button to create another Batch List (following steps beginning on page 3 of this section, Setting Up a Batch List).
   b. Load Batch Lists:
      i. Load plates into the OmniLog as shown in the plate stack. (Biolog imprinting facing toward you while loading.)

ii. OmniLog must not be actively reading plates. You will need to wait until any current reads are done.

iii. Click on the Move Camera button if you need to place MicroPlates into the tray at the current camera position.

iv. After all MicroPlates have been loaded (for all pending Batch Lists), click the Load Batch Lists button.

v. The incubation time clock will now begin for the Batches loaded.

c. Clear Batch Lists:

i. Click Clear Batch Lists button to clear all pending Batch Lists.

2. If you have loaded your Batch Lists or cleared any pending Batch Lists, the Read window will be displayed.

3. The Read window displayed will show any new Batches that were loaded.

4. All MicroPlates in a batch will have the same setup time.

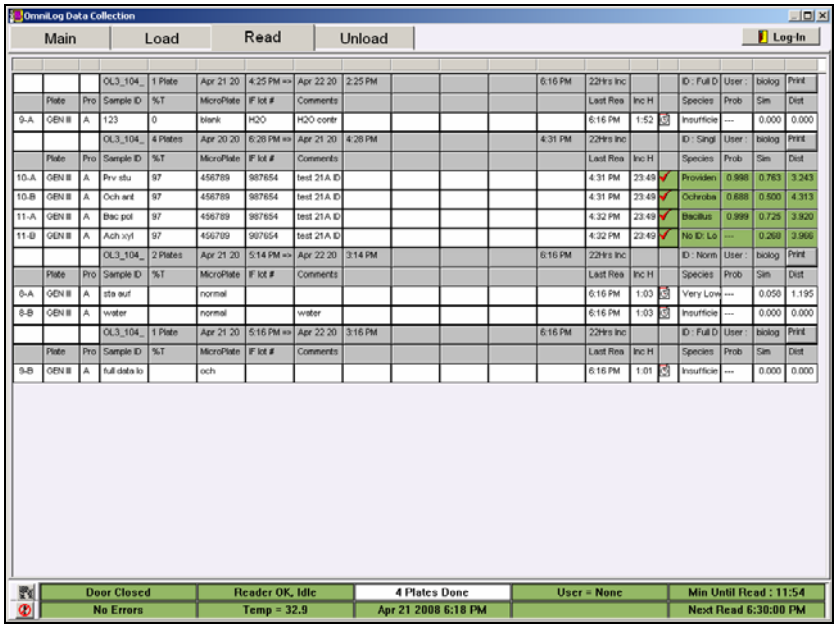


Figure 29 Read Window (with entries)

Rules of thumb for loading batches

Assess Batch

- Does my batch contain MicroPlates that are difficult to set up?
- Do I want to read in different read modes?
- Am I a beginner at preparing samples and inoculating MicroPlates?

If the answer to these questions is yes, break batch up into several batches (approximately 5 MicroPlates per batch).

Assess Stack Status

Before opening the door, check the Load Plates window, which shows the status of each stack slot.

A or B	⇒ Left (A) or right (B) stack column
Entry in each box	⇒ Gives sample identifiers
Red  (with padlock icon)	⇒ Slot occupied by MicroPlate not yet unloaded. Do not use
Green  (with hand icon)	⇒ OK to use this slot

Open Door

- Open the incubator/reader door

Remove Tray and Insert MicroPlate(s)

- Slide one tray out of its slot
- Place tray on bench top
- Insert 1 or 2 MicroPlates into that tray
  - ⇒ Make sure “Biolog” logo faces front of tray (toward you)
  - ⇒ Make sure A1 well is in right rear corner
  - ⇒ Make sure MicroPlate fits into slot and all four corners are well seated
  - ⇒ Make sure MicroPlate lid is securely in place

Replace Tray in Incubator/Reader

- Slide the tray back into its exact slot in the incubator/reader

  ⇒ Make sure you do not put tray into wrong slot

If you do put tray in the wrong slot, the system will read it and identify it anyway, but the identification will not match that correct sample identifiers. OmniLog ID software cannot detect this kind of error. This depends on whether the slot is entered in a batch.

Repeat for all MicroPlates

- Remove next tray to be loaded
- Place it on bench top
- Insert 1 or 2 MicroPlates into that tray
- Load into incubator/reader

Watch the time!

- Do not allow MicroPlates to sit on bench top for more than 20 minutes

Close Door

- Close the incubator/reader door

Moving the camera when loading MicroPlates

In the normal course of operation, the camera in the incubator/reader moves from slot to slot as it reads MicroPlates. When not in use, it usually "parks" itself at the top row of the stack (slot 25). Occasionally it will park at slot 1. Wherever it parks, that tray locks into place; you will not be able to remove it for loading. If, for example, if the incubator/reader is empty and you are loading a batch of 50 MicroPlates, you will not be able to remove tray #25 until you move the camera out of the way. Occasionally the camera will move behind a slot while you have that tray out to load MicroPlates. In both cases, you must move the camera out of the way.

- If you encounter any difficulty inserting a tray all the way or closing the door because one tray is sticking out, it is most likely because the camera has locked that slot.
- DO NOT FORCE TRAYS INTO SLOT
- Simply remove the tray and move the camera out of the way using OmniLog ID software

To move the camera:

- Remove the tray causing the problem
- Close the door
- Click the Move Camera button on the Load Plates window

Note: If you click the Move Camera button before closing the door, you will get an on-screen prompt to close the door.

- The camera will move to another slot. An on-screen message will appear, telling you when it's ok to open the door and resume loading that row
- Open the door and load the tray

How often do readings take place?

- The OmniLog ID reads every 15 minutes
- These intervals occur every 15 minutes (at every quarter hour) on the computer clock, not 15 minutes from the time the MicroPlates are loaded
- Only MicroPlates that fall within these guidelines will be read

What are OmniLog ID's identification criteria?

An OmniLog ID is made when two consecutive readings with acceptable SIM values have the same ID call (exception: Immediate read).

Normal mode read:

- An ID made any time during the 22 hour read window will be saved
- Reading will stop and the MicroPlate is mark Done with a green check mark
- If no ID occurs in the 22 hour window and there are too few positive reactions then the readings will be extended to 36 hours until a final ID call is made

Full Data Logger mode read:

- If an ID is made any time during the 22 or 36 hour reads, an ID printout report will be generated
- Reads will continue for the full 22 or 36 hours user defined setting

Immediate (single) mode read:

- ID is made on a single reading with acceptable SIM values

Section 7. Unloading MicroPlates

Removing MicroPlates from the incubator/reader requires only a few easy steps:

1. Check the status of all MicroPlates to see which ones are ready to be unloaded.
2. Make sure that IDs have printed out.
3. Remove MicroPlates that are ready for unloading.
4. Check status again.

Checking Unload Status

When MicroPlates are ready to be unloaded, the footer bar will let you know by reporting "X Plates Done" (X = the number of MicroPlates finished). During operation, the Read window will be displayed during incubation and reading MicroPlates. The Read window allows you to view the batches and the MicroPlate status.

Once you open the door, you have a limited time to load and unload MicroPlates. If you need more time, activate the Snooze bar

See Section 9.

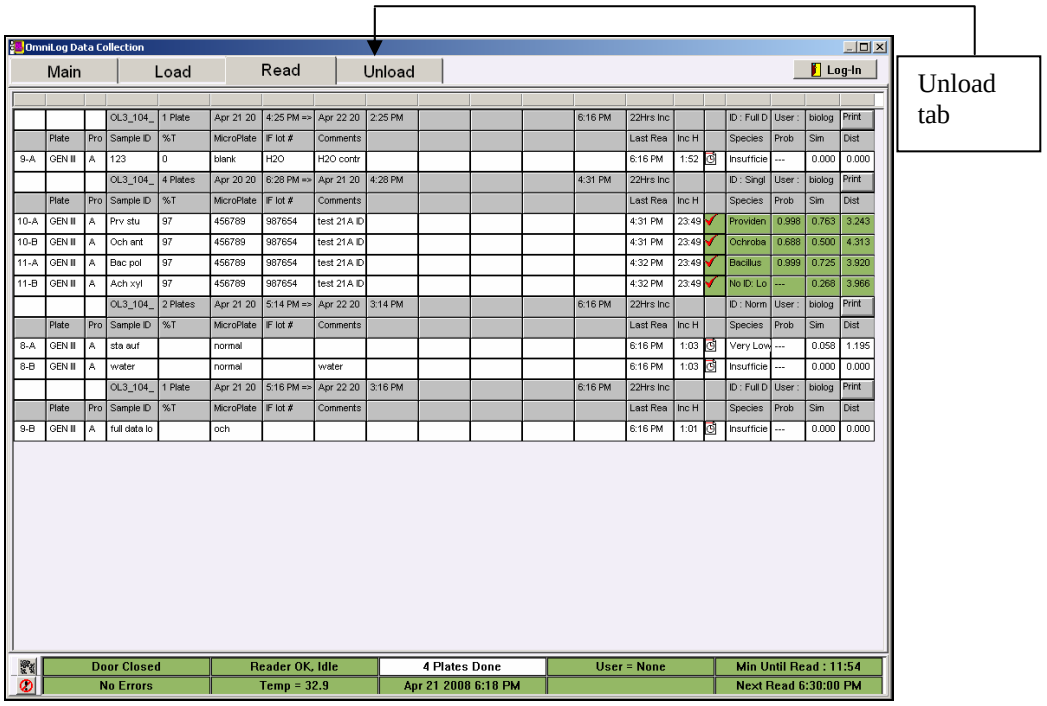


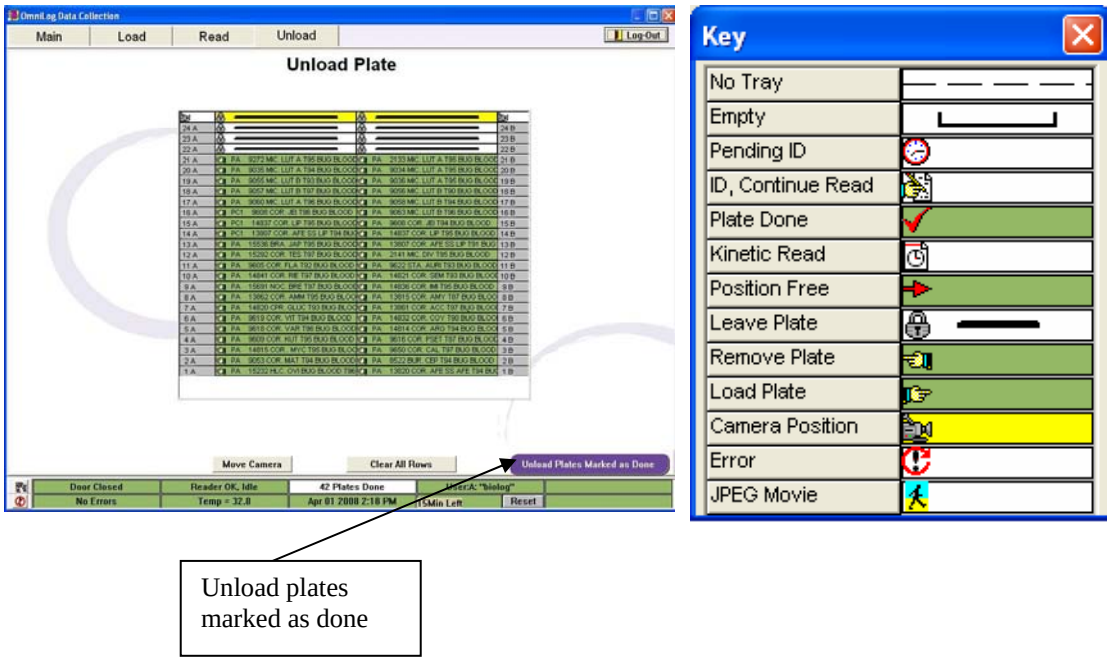
Figure 30 Read Window

1. When the **Read** window indicates that some MicroPlates are ready to be unloaded, click **Unload** on the top menu bar.
2. The **Unload** window shows a picture of the tray stack, indicating current status (see Table 9 for explanation of symbols).
3. You can click on any row to see the **Unload Key**. Check to ascertain which MicroPlates are ready for removal.

Table 9 Unload Window Symbols

Key	Explanation
Numbers along left and right edges	Numbers correlate to tray numbers, starting with number 1 at the bottom through number 25 at the top
A and B designations along left and right edges	A = left column of MicroPlates B = right column of MicroPlates
White slots with ----------	Slot empty of both MicroPlates and tray
White slots with [_____]	Slot contains tray, but not MicroPlate
Green background with red checkmark icon	Slots that contain MicroPlates that have been read and are ready to be removed
White background with hand writing icon	Slots containing MicroPlates that have been read but are not ready to be removed. Data still saving.
White background with clock icon	Slots containing plates that have not been read yet

Table 10 Unload Window (View as Plates) and Load Key



Removing Done MicroPlates

Caution!

Do NOT open the incubator/reader door when the footer bar shows the message "Reader Busy."

Removing MicroPlates not listed as "Done" on the Unload Plates window cannot be detected by the system.

1. The OmniLog must not be actively reading plates. Wait until any current reads are done.
2. Unload plates from the OmniLog as shown in the plate stack.
3. Remove only one tray at a time.
4. Slide the tray completely out of the incubator/reader.
5. Place the tray on the workbench.
6. Remove the appropriate MicroPlate(s) from the tray.
7. Put the tray back into the same slot in the incubator/reader.
8. If the camera has parked itself into slot behind a tray you wish to remove and you cannot get the tray out, click **Move Camera** and select another row.
9. After all MicroPlates have been unloaded (for all completed MicroPlates), click the **Unload Plates Marked as Done** button.
10. After you've removed all the desired MicroPlates, close the door.

Removing All MicroPlates

Clear All Rows:

11. Click on the **Clear All Rows** button to clear all MicroPlates loaded (done and not done).
12. You will need confirm the request to “clear all rows”.
13. You must have edit privileges to use this function.

Marking MicroPlates As Done

Note: To Mark MicroPlates as Done, Users must minimally have Edit Privileges.

During normal operation, the Read window will display. As you track the progress of readings, you may occasionally detect, in your judgment, a clearly erroneous identification by the OmniLog incubator/reader. This may occur if a sample has been mis-prepared, a MicroPlate incorrectly inoculated, incorrectly labeled or places in the tray backwards.

Rather than proceed with a final reading of that MicroPlate, you can clear it from its batch. To clear a MicroPlate record during the read process in Restricted Access mode:

1. Click on the appropriate row in the **Read** window.
2. The **Plate Data** window appears.
3. Click **Mark as Done**. That entry will now be marked as "Done" on the **Read** window listing.
4. To restore the entry, click **Restore**.
5. Close the window by clicking the windows **X** (upper right corner) or **Done** button (lower left corner).
6. The software will return to the **Read** window.

Special incubation time change function:

There is an option on the Plate Data Page to change the incubation time. The User may wish to use this for example if they decided to change from 22 hours incubation to 36 hours incubation in full data mode for the development of a User database.

Plate Data Window Print functions

Print buttons:

1. **Print Screen**- Prints the current display on the screen (default is portrait, change to landscape to get printout with complete view).
2. **Print Report**- Only available if the ID result is final.

3. **Print Preview**- Displays the final report with the following options:
   a. **Close**- Click to close the **Print Preview** screen.
   b. **Print**- Click to print the final ID report in view. (Note: If while the Print Preview display is up, another read process occurs and another final ID call has been made, the click of this **Print** button will not print the report in view. This Print button will instead print the last final ID report sent to the printer).
   c. **Select Printer**- Click to get to Windows **Select Printer** dialog.
   d. **Printer Setup**- Click to access Windows **Printer Setup** dialog.
   e. **Page Setup**- Click to access Windows **Page Setup** dialog.

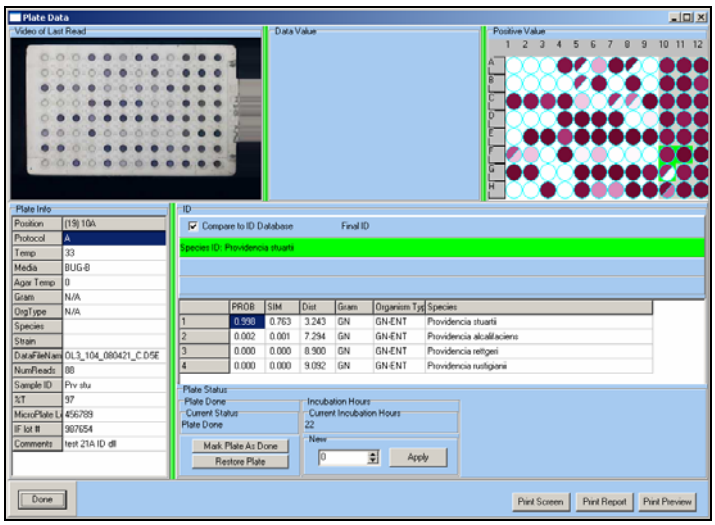


Figure 31 Plate Data Window

Understanding Naming Conventions

Before using the advanced data management functions available in the RetroSpect Trending and Tracking data management software, it is important to understand how batches and data files are named. File names and lower storage path levels are created automatically. The file names are keyed to the project code defined by the user, setup date of the batch and the batch code letter. The batch code letter is assigned by the software. The storage path is by project code and further by the year and month.

Table 11 shows these conventions.

Table 11 File Name and Pathway Naming Format

File Type	Automatic Coding	Example
Path	Path: …..\PPP\YYYY\MM\ = \ 3 postion alpha numeric project code\4 digit year\2 digit month	IDS project, data for March, 2008 = C:\.....\IDS\2008\03\
File name	3 postion alpha numeric project code_3 digit OmniLog SN_ YYMMDD_+ single letter (batch identifier) + ".D5E"	Data for March 13, 2008, project code IDS, OL serial number 101 = IDS_101_080313_A.D5

Section 8. Interpreting Results

During normal operation, the **Read** window will display. As OmniLog Data Collection makes identification calls, the results will show for all MicroPlates in all current batches. The batches are shown in the order in which they were loaded, with the one that's been in the longest on top. Scroll down to see all results.

Each batch begins with a two-row header.

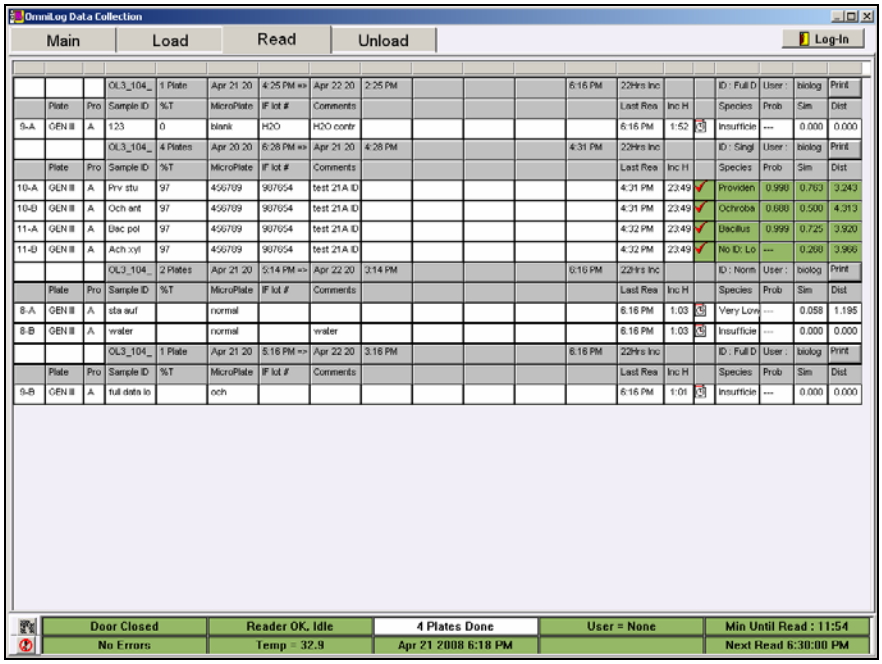


Figure 32 Read Window with Results

Table 12 and Table 13 explain the column headings on the Read window. Expand column widths to view complete text.

Table 12 Read Window Columns (top row)

Column Heading	What It Means
NP4_110_080328_F	Project code/OL serial number/ yy/mm/dd/ batch code
41 plates	Number of plates in a batch
Mar 28 2008	Start date
11:56 AM⇒	Start time
Mar 29 2008	End date
9:56 AM	End time
4:32 PM	Last read
22 hrs Inc	Time set for incubation
ID:	Indicates the read mode selected
User:	Indicates the User setting up the Batch
Biolog	Indicates the Database for search
Print	To print the final report, if necessary.

Table 13 Read Window (second row)

Column Heading	What It Means
Blank field	The designated tray position
Plate	The type of MicroPlate used for that sample
Protocol	The setup protocol used
Sample ID (Field Name 1 default)	User Field 1 information
Field Name 2	User Field 2 information, if required
Field Name 3	User Field 3 information, if required
Field Name 4	User Field 4 information, if required
Field Name 5	User Field 5 information, if required
Field Name 6	User Field 6 information, if required
Field Name 7	User Field 7 information, if required

Table 13 Read Window (second row)

Column Heading	What It Means
Field Name 8	User Field 8 information, if required
Field Name 9	User Field 9 information, if required
Field Name 10	User Field 10 information, if required
Last Read	When MicroPlate was read last
Inc. Hrs	How long the MicroPlate has been incubating
ID Status indicator	Shows whether result is in-progress or final • green box with red checkmark means final ID • white box with clock face means in-progress ID or not read yet • ! with 'international no symbol' means "Missing Plate' • in-progress results are updated every 15 minutes, at each reading
Species ID	Gives full identification of microbe when the ID status indicator shows a final ID call. Preliminary call status may appear as the reading period is progressing; no display shall be considered final until the ID status indicator column indicates a final ID call.
Prob	Confidence probability of a called ID
Sim	ID Score value
Dist	Equivalent number of mismatches

Possible ID Call Results

- Species ID
- Genus ID
- NO ID: Low SIM
- NO ID
- NO ID: False Positive
- No ID: Too Many Borderline Reactions
- NO ID: Insufficient Pattern

Note: The headings on the **Read** window batches reflect the information you selected during batch setup. As a result, these headings will change depending on your data entered.

1. Check the **Read** window for in-progress and final identifications.
2. For more detailed results click on any part of any row in the **Read** window. A **Plate Data** window will appear for that MicroPlate.
3. The Identification is not called yet on the **Read** window unless there is a check mark with a green background in the plate status column of the Plate position row.
4. Preliminary result displays will change as the incubation and reads progress.
5. If you get a final "No Identification" result, you can check mismatches on the **Plate Data** window. The left top color image represents threshold reaction call of the test MicroPlate. The lower right color represents reference database pattern for the most closely matched species. (Viewing and printing the Threshold Value information is an optional setting, set by the Program Administrator).
    a. If mismatches are mixed with some wells positive and others negative with respect to the closest match pattern, your test strain may not in Biolog's database. You can add it to your own user-created database (using RetroSpect Trending and Tracking software).
    b. If the mismatches are all positive or all negative, you may have made a testing error. Refer to Table 14 to assist you in figuring out the cause of these mismatches. Or see Trouble shooting Section 12.
6. The Read window gives the first ID choice for each microbe. To see the top 4 choices for a Final ID call, click any row on the Read window. The first four choices will show in the lower section of the Plate Data window. (Viewing and printing the ID List is an optional setting, set by the Program Administrator).

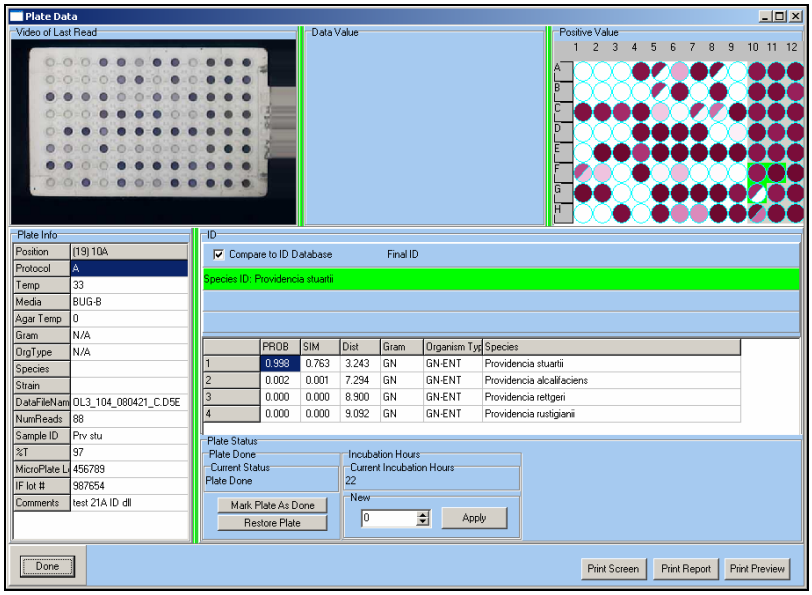


Figure 33 Plate Data Window

Table 14 Assessing Mismatches

Mismatch Type	What It Might Mean
All mismatches + for reference pattern vs – for test pattern. (test pattern is giving fewer positive reactions than the species you're comparing it to)	Under-inoculation
	A1 well is overfilled, contains clumps, or is cloudy
	Cells were mishandled, too old, cultured on the wrong medium, suspended in wrong inoculating fluid, incubated at wrong temperature.
All mismatches – for reference pattern vs + for test pattern. (test pattern is giving more positive reactions than the species you're comparing it to)	Over-inoculation (especially with enterics)
	A1 well is underfilled
	Contamination (mixed culture)

ID Statistics Background

The OmniLog 2.X series software measures the validity of the ID based on two criteria: "Match" and “Separation". A value is derived for both of these concepts based on the Distance (DIST). The DIST value indicates how many mismatched reactions the test organism has to the reference organism in the database. "Match" is a measure of how good of an absolute match the data is to the choices selected in the database. The smaller the DIST, the greater the match. “Separation" is the difference in distance between the first choice and all of the other choices. These two values are then multiplied together to get a final net ID score. That final number is called "SIM" in the MicroLog and OmniLog systems.

The match score itself constantly decreases with increasing distance. The value for separation by itself is shown as "PROB" when an ID is called. If the fist choice and the second choice were the exact same distance from the entered data, then the PROB would be 0.5. If the first three choices all had the same distance, the PROB for all three would be 0.3333, and so on. As the differences in the DIST gets larger, the PROB goes up. At a difference in DIST of 1.0, the PROB is 0.950. At a difference in DIST of 2.0, the PROB is 0.997.

If a well result does not match the result in the database for the organism in question plus/minus mismatch indicators are visible on the screen (upper left vs. lower right color interpretation) and on the printout for the original read for that well. If the read is positive on the MicroPlate, <X>, and the database result for that well is negative the printout shows <X- to indicate a mismatch where the database reaction is negative. If you have a negative read, X with no brackets and database value for that well is positive the well will read X+ indicating a positive reaction in the database. At the time of a read the data is compared to the database to determine the ID.

ID Call Criteria

SIM cut-off

- The SIM cut-off for an identification call is a sliding scale based on the hour
- It is 1.0 at 0 hours, and goes down linearly to 0.5 at 8 hours
- It is 0.5 for any time longer than 8 hours
- The SIM is "over" the cut-off if it is greater than or equal to the cut-off

ID call

- The ID is called after two consecutive reads where the SIM gets over the cut-off with the same ID call. (Immediate (single) read mode: call on one read.)
- The Reading of the plate will stop when and ID has been made
- The reading of a plate in Normal ID mode will stop when the first of the following happens:
  - An ID has been made
  - The incubation time has been reached
  - The plate turns false positive

ID Result Calls

The following table (Table 15) shows the ID Results Call as a single line that appears as the final ID Result in the printed report.

Table 15 Printed final Report Possible ID Calls

Final ID Call	What It Means
Species ID: + First Choice Species Name	If the plate has a first choice SIM that exceeds that threshold in two consecutive reads. Immediate read mode uses only one read for an ID call.
Genus ID + Genus name	On the final read (22 or 36 hours), Biolog will give a genus-level ID rather than a species-level ID if all of the following is true: (a) SIMs for top IDs are < 0.50 (b) all belong to the same genus (c) if their total is > 0.50
No ID	ID Searched, SIM too low
No ID: Low SIM: + First Choice Species Name	If the plate is on the final read and has all of the following: (a) 2 or more metabolic positive reactions (b) the first choice species has matched a previous read (c) a first choice SIM that is greater than or equal to 0.2 but below the ID call threshold.
No ID: False Positive	The following are considered an unacceptable level of False Positive and the reading of this plate is discontinued. (a) If the Raw OL A1 value is >= 225 (b) If the Raw A1 value is >= 125 and the current number of positives is more than 5 below the maximum number of positives for the plate so far
No ID: Too Many Borderline Reactions	A too many borderline call will be made if there are >25 borderline reactions on the plate.
No ID: Insufficient Pattern	A insufficient pattern call will be made if the plate has either: (a) fewer than 2 positive metabolic reactions, or (b) the first choice ID does not match the previous read first choice ID, or (c) the first choice SIM is less than 0.2.
Missing Plate	Software detects that the plate is missing.

ID Notice

Table 16 shows advice for further testing on **No-ID** calls will be provided on the Final Report printout.

Table 16 Printed ID Notice Possibilities for No ID Calls

Current Protocol	Final ID Notice	What It Means
A	Retest with Protocol C1	No ID and Insufficient Positives
B	Retest with Protocol A	No ID and Insufficient Positives
C1	Retest with Protocol C2	No ID and Insufficient Positives
B	Retest with Lower Inoculum (T 98%) or Protocol B	No ID and False Positive
C1	Retest with Protocol A	No ID and False Positive
C2	Retest with Protocol C1	No ID and False Positive

Read Window ID Display

Table 17 shows the one-line display at Read menu, used as a status report on plates being read. Note: All results are preliminary and may not be indicative of the final result until the status box indicates with a checkmark on a green background that the result is final.

Table 17 Read Window ID Display Possibilities

Read window display	What It Means
Not Read	-Not Read Yet
Insufficient Pattern	-Not Final read, Insufficient Pattern
Incubation Extended Past 22 Hours: Insufficient Pattern	-Insufficient Pattern, 22 Hour incubation time reached
No ID: Insufficient Pattern	- Insufficient Pattern, Incubation time-out (Protocol C2)
No ID: Insufficient Pattern: + ID notice	- Insufficient Pattern, Incubation time-out (All other protocols.)
Very Low SIM	- Not final read, Sim < 0.2
Too Many Borderline Reactions	- Not final read. too many borderlines
No ID: Too Many Borderline Reactions	- Time-out too many borderlines
Low SIM:" + species name of first choice	- Not final read, Sim >= 0.2, < cut-off
Species name of first choice	- ID Called
No ID: Low SIM:" + species name of first choice	- Time-out, ID searched, Sim >= 0.2, < cutt-off
No ID	- Time-out, ID searched, Sim < 0.2
No ID: False Postive	- False Positive (Protocol A)
No ID: False Positive: + ID notice	- False Positive (All other protocols.)

Section 9. Special Functions

OmniLog ID has several special functions that allow you to do the following:

- Get a little extra time before the next reading.
- Remove a MicroPlate from a batch if an error has occurred in the preparation of that MicroPlate.
- Perform an immediate read if you have incubated MicroPlates in a secondary, offline incubator.
- Exit the software in special circumstances.

Using the Snooze Bar

At times you may wish to gain a few extra minutes before the next reading. If, for example, the incubator/reader door is open and you are still loading and/or unloading MicroPlates, the footer bar may begin to blink and say "Next Read 5 Min." Just below that entry, the **Snooze** bar will appear indicating that you have 2 or less minutes until a read is performed.

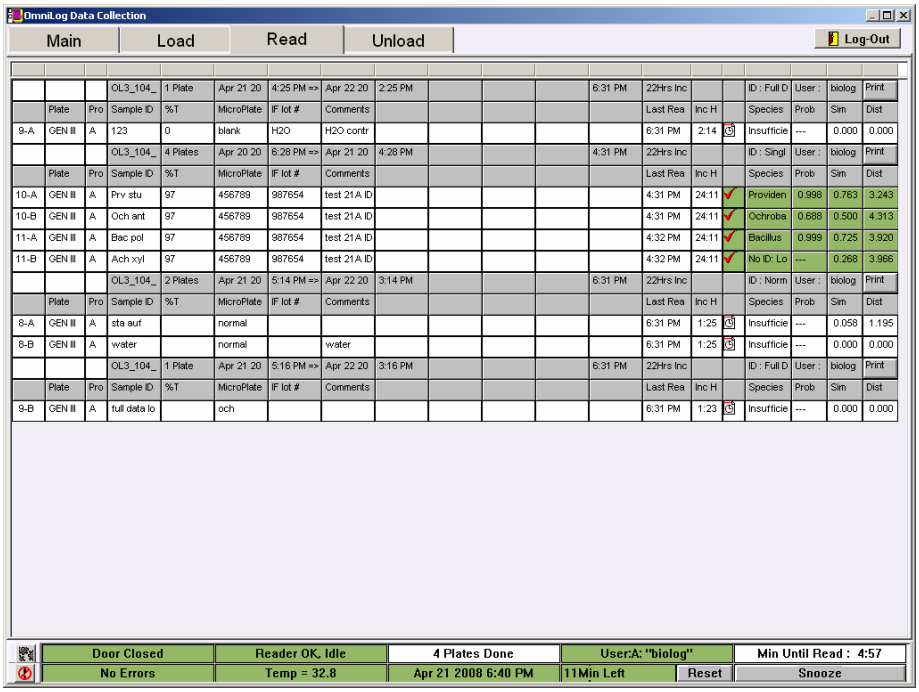


Figure 34 Read Window with Snooze

To gain extra time:

1. Click **Snooze**.
2. The **Snooze** bar will disappear. It will add 2 more minutes before a read will begin.
3. You can continue to click the **Snooze** bar to delay the read several times.
4. If you try to click **Snooze** after a first skipped read, a pop-up window will ask "Are you sure you want to do that?"
5. These pop-up queries will time out after 2 minutes.
6. If you try to snooze too close to the read time the software will not allow the snooze and will immediately read the plates.
7. If you have walked away with the door open, a message will appear saying "Door Open." An alarm will sound.

Using an Offline Incubator

At times you may want to use a second incubator, then place the incubated MicroPlates into the incubator/reader for immediate reading. This is especially helpful in the following circumstances:

- If your suspected microbe requires a special atmosphere, such as 6% CO2
- *If your suspected microbe requires incubation at a temperature different from the current incubator/reader temperature, and you do not wish to reset the temperature

1. Prepare samples according to Section 5.
2. Incubate MicroPlates in an offline incubator for the appropriate length of time.
3. Follow the **Load** process described in Section 6. Click the **Create Batch List** button.
4. On the **Batch Setup** window, make sure you click the **Immediate Read** radio button.
5. Enter your **Project code**.
6. Check that the **ID Database to Search** is correct.
7. Click the **Next** button.
8. Click **Edit Entry** button to get to the **Plate Information** window.
9. Enter sample information on the **Plate Information** window, as described in Section 6.
10. Proceed with loading and reading as described in Section 6.

11. OmniLog ID will immediately read MicroPlates marked as "Immediate Read".

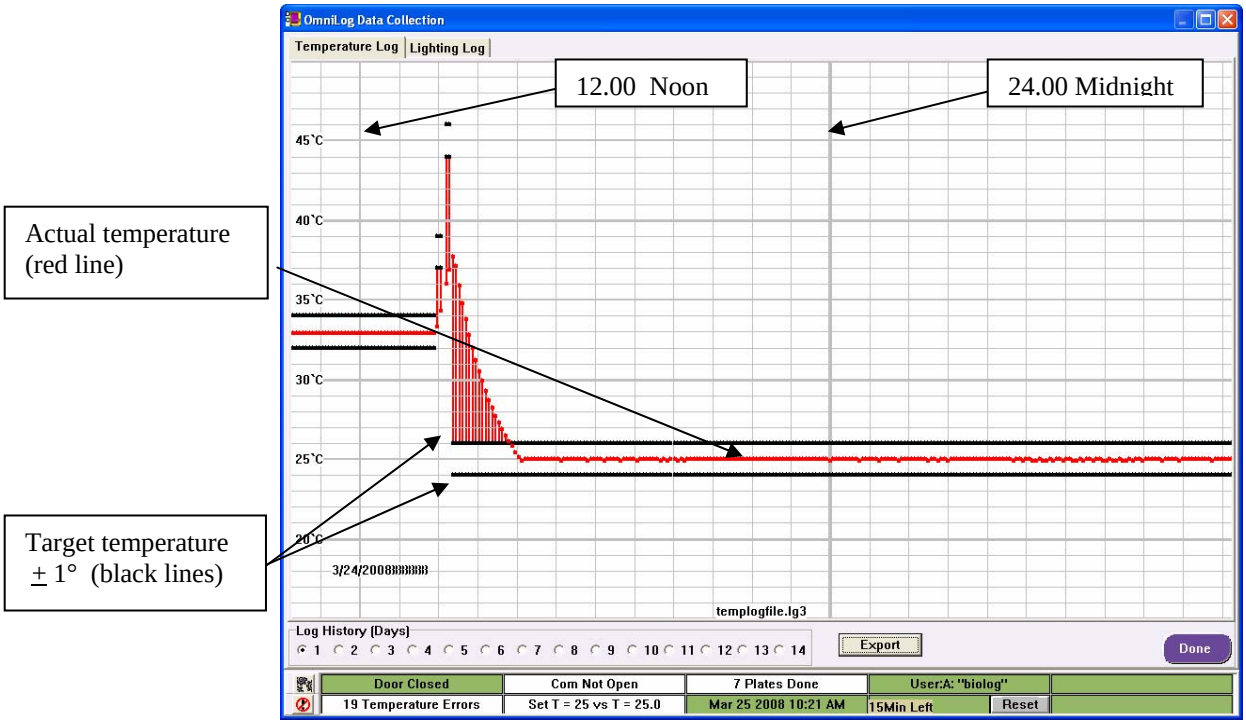


Figure 35 Temperature Log Window

Temperature Log

1. Click on the **Setup** button on the **Main** window.
2. Click **Temp/Lighting Log** button to access a graphical log of 1-14 days of actual temperature readings for the incubator/reader.
3. With the **Temperature Log** tab selection, the **Temperature Log** appears, displaying black horizontal lines that represent the target temperature range (1(less and 1(more than set value). Figure 35.
4. The red line represents the actual temperature record.
5. Each vertical line represents one hour.
6. The medium thickness line represents noon and the thick line represents midnight.
7. To save a file of the temperature readings, click the **Export** button.
8. Using the Window **Save As** dialog, save the file as a csv file in the location of your choice.

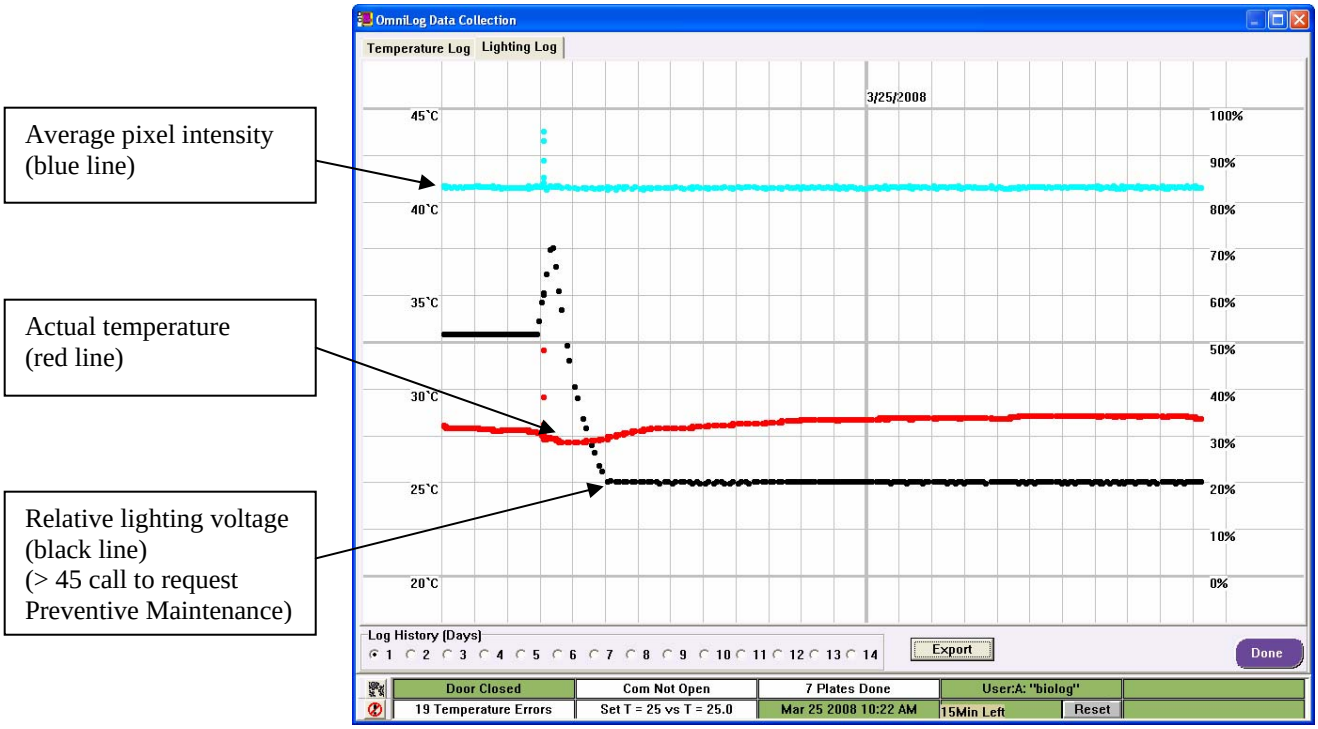


Figure 36 Lighting Log Tab Window

Lighting Log

At times in order to trouble shoot video problems, Biolog Technical Services personnel may instruct you to capture the information provided by the Lighting Log.

1. Click on the **Setup** button on the **Main** window.
2. Click **Temp/Lighting Log** button to access a graphical log of 1-14 days of actual Light values for the incubator/reader.
3. With the **Lighting Log** tab selection, the **Lighting Log** appears. Figure 36.
4. The blue line represents the average pixel intensity as set in the factory (scale on the right).
5. The red line represents the actual temperature (scale on left).
6. The black line represents the relative lighting voltage (scale on right).
7. Each vertical line represents one hour.
8. The medium thickness line represents noon and the thick line represents midnight.

9. To save a file of the lighting values, click the **Export** button.
10. Using the Window **Save As** dialog, save the file as a csv file in the location of your choice.

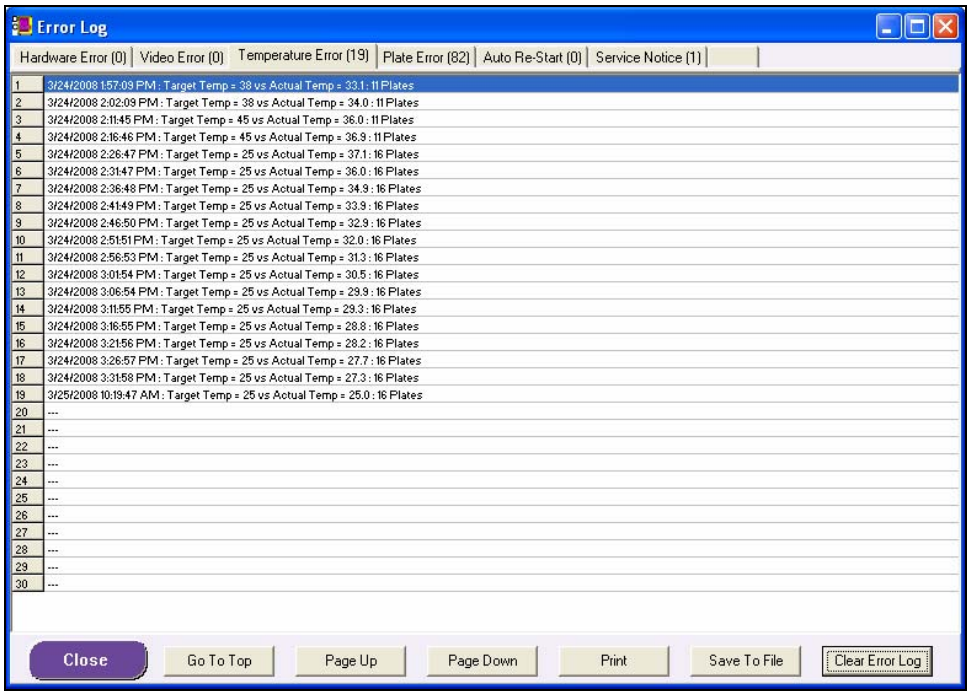


Figure 37 Temperature Error Tab in the Error Log Window

Error Log

The Error log can be accessed two ways.

1. In the **Setup** window, click the **Error Log** button in the **Log** section.
2. Click on the footer **Error status** bar and the **Error Log** will open.

The Error log provides information under the following categories.

Hardware Error	Detects problems with the Hardware such as tray jams and power interruptions.
Video Error	Detects any problems with the capture of the image by the Frame grabber (Video card) or problems with the camera.
Temperature Error	Detects each instance where the target temperature and the current temperature are not within allowable tolerance when batches of MicroPlate are being read.
Plate Error	Detect plate errors such as: detecting a missing plate within a loaded batch list.
Auto-Re-Start	Informs the user when an Auto Re-Start occurred (e.g. during a power outage).
Service Notice	Notices to provide information to service technicians, these do not impact the system performance.

Navigation options for each of the tabs.

Go to Top	Click to navigate to first entry.
Page Up	Click to move a single page view, to earlier entries.
Page Down	Click to move a single page view, to later entries.
Print	Click to print the entries with your installed printer.
Save to File	Click and using the Window Save As dialog, save the file as a text file in the location of your choice.
Clear Error Log	Click to clear completely all entries in the current tab in view.

Relocating the OmniLog

Caution!

Once the OmniLog ID system is set up, it's best NOT to move it. If for some reason you must move it, call Biolog Technical Services first, then follow these instructions.

If you physically move the OmniLog, you must first park the camera, as follows:

1. On the **Main** window, click **Setup**. Make sure the com port is open, the reader is initialized, and the cycle mode is off.
2. Move the camera to the bottom of the reader, using the **Move Camera** buttons. The normal resting position for the camera is row 1 (Park).
3. Wait until the footer bar Reader Status cell goes from **Busy** to **Idle**.
4. Quit the OmniLog software and turn off the OmniLog computer.
5. It is advisable (for weight considerations) to remove all other trays from the incubator/reader before transporting it.
6. Remember to re-install all trays before using it again.

Quitting the Software

Caution!

Quitting the software while there are pending reads will cause you to miss readings. Except when data loss is unavoidable, make sure the software and incubator / reader are on while there are pending MicroPlates reads.

Since the computer you purchased with the system is dedicated to the OmniLog Incubator/reader, you should not run any non-Biolog software on it.

If you do need to quit the software to move the OmniLog Incubator/Reader to another location, the software will save all current MicroPlate information and data to files. When you start the OmniLog software back up, the software will retrieve all the MicroPlate information that has been saved. It will resume all pending reads, using the current time as the read time (not the plate setup time). If you quit the software while readings are pending, you will lose all the readings that would have occurred while the system was off.

1. Click **X** on the Windows commands (upper right corner of the window).
2. A “pending read” status warning will appear in the center of the screen. If there are pending reads, the warning box will be red; if not, the box will be green and the message will say "OK to exit."
3. . A User must minimally have Edit privilege to exit the software.
4. Click Exit. The OmniLog Data Collection Software will close.

Figure 38 Exit Window

Section 10. Technical Notes

This section gives specific information regarding consumable materials you'll need to perform microbial identifications, as well as instructions for how to prepare isolation media and inoculating fluid.

Materials List

Table 18 Materials List for the GEN III Protocol

	Item	Description
Media	BUG + B	Growth medium for gram-negative and gram-positive bacteria
	BUG	Growth medium for agricultural bacteria
	CHOC	Growth medium for fastidious gram-negative bacteria
Miscellaneous Supplies	Streakerz™	Sterile 6” pointed streaking sticks
	Inoculatorz™	Sterile Inoculatorz cotton-tipped swabs
	Transfer pipettes	Sterile 9” disposable pipettes, graduated tip, 5 ml
	Reagent reservoirs	Sterile reservoirs
	Pipette tips	Sterile, racked, for repeating multichannel pipetter
MicroPlates	GEN III MicroPlates	MicroPlates for aerobic bacteria

Media Preparation

Biolog can provide you with all the pre-made media you will need for microbe identification. However, you can also prepare your own agar media, using the Biolog dehydrated medium formulas.

Making Biolog Universal Growth Agar + blood (BUG + B)

Caution!

If you're making your own media, follow instructions exactly. Take care to avoid contamination.

1. Mix the following in a 2-3 liter container:
   a. 57 grams BUG Agar
   b. 950 ml purified, distilled, or deionized water
2. Gently boil mixture while stirring to dissolve the agar and other components.
3. Cool an aliquot and measure the pH. Adjust pH with NaOH or HCl to achieve a final pH of 7.3 + 0.1 at 25(C.
4. Sterilize by autoclaving at 15 pounds of pressure and 121(C for 15 minutes.
5. Cool to 45-50(C.
6. Add 50 ml sterile fresh defibrinated sheep's blood just prior to dispensing and mix gently. (Use good quality blood with a hematocrit of at least 40%).
7. Dispense into sterile petri dishes.

Making Biolog Universal Growth Agar (BUG)

1. Mix the following in a 2-3 liter flask:
   a. 57 grams BUG Agar
   b. 1,000 ml purified, distilled, or deionized water
2. Gently boil while stirring to dissolve the agar and other components.
3. Cool an aliquot and measure the pH. Adjust pH with NaOH or HCl to achieve a final pH of 7.3 + 0.1.
4. Sterilize by autoclaving at 15 pounds of pressure, at 121(C, for 15 minutes.
5. Cool to 45-50(C.
6. Dispense into sterile petri dishes.

General hints for culture media preparation

Keep dehydrated media powder in original bottles with lids tightly closed to avoid water absorption and deterioration.

Use clean dry glassware that has been rinsed free of all soap residue.

Add water to the vessel first, then weigh agar powder and add it to the vessel. Mix to obtain an even suspension. Do NOT fill the vessel more than two-thirds full (to avoid boiling over during heating and autoclaving).

Heat agar gently, with constant stirring.

Inoculum Preparation Hints

General Hints For Inoculum Preparation

1. For extremely clumpy bacteria that cannot be dispersed directly, use the following procedure.
    a. First prepare a dense suspension in 2 ml of IF as follows.
        i. Use a sterile wooden Streakerz stick to remove a clump of cell mass from the agar surface without gouging the agar.
        ii. Use the Streakerz stick to crush, break up, and spread the clumps of cells against and along the inner wall of the tube.
        iii. Then add 2 ml of IF, and gradually slide the dispersed cells into the IF.
        iv. The resulting cell suspension will be a mixture of suspended cells and residual clumps.
        v. Stand the tube in a rack for about 5 minutes and allow the clumps to settle to the bottom.
        vi. Use a small pipet and transfer the suspended cells at the top into afresh tube of IF to achieve the target cell density.
2. Pure cultures must be used to obtain identifications.
    a. The system is not designed to identify individual bacterial strains from within mixed cultures.
    b. The most common problem in identification is that microbiologists are not aware that they have a mixed culture.
    c. Streaking for isolated colonies may not be sufficient because isolated colonies can arise from a mixed clump of cells as well as a single cell.
    d. Bacteria have sticky surfaces, and they tightly adhere to other bacteria. This is particularly a problem with mucoid bacteria, fresh environmental isolates, and staphylococci.

e. First, examine cultures with care using a dissecting microscope or some colony magnification to make sure that only one colony morphology is present in the culture.

f. If no species identification is obtained, it may still be a mixed culture.

g. Restreak the cells onto a multi-chromogenic agar medium and let the original agar plate and the chromogenic agar plate sit at room temperature for 3 or 4 days.

h. Examine both plates carefully, looking for the outgrowth on "bumps" or non-uniform growth in the areas of confluent growth.

i. On the chromogenic agar plate, look for more than one color.

j. If necessary, re-isolate the colony types that are present and perform the identification assay a second time.

3. Culture media and repeated subculturing may affect the results. Strains may produce different phenotypic patterns depending upon how they are cultured prior to inoculation.

4. Sterile components and aseptic techniques must be used in set-up procedures. Contamination will affect results.

5. Disposable glassware should be used to handle all cell suspensions and solutions. Glassware that has been washed may contain trace amounts of soap or detergent that will affect results.

6. Prewarm the IF and the MicroPlates to room temperature before use. Some species (e.g., Neisseria sp.) are very sensitive to cold shocks.

7. Check the calibration of the turbidimeter carefully and always prepare your inoculum within the specified density range.

8. Biolog's chemistry contains components that are sensitive to temperature and light. Store the inoculating fluids in the dark with refrigeration. Brown or yellow wells in the GEN III MicroPlate indicate deterioration of the chemistry.

9. Always keep in mind that you are testing the metabolic properties of live cells. Some species can lose their metabolic vigor when subjected to stresses (e.g., temperature, pH, and osmolarity) for even a few minutes. To get the best performance possible from these MicroPlates, be aware that the cells are alive and take care in how they are handled.

Section 11. Frequently Asked Questions

Q. How I should I store my MicroPlates?

A. Unpack MicroPlates as soon as you receive them and keep them refrigerated until use. Biolog MicroPlates should be kept refrigerated (not frozen). MicroPlates are fairly stable at room temperature; refrigeration requirement are to obtain maximum shelf life. At moderately warm temperatures, they slowly begin to deteriorate. Visually examine MicroPlates before using them. You may see faint yellow-brown or pink shades in wells when they are dry (this is OK). However, if wells show significant color immediately after inoculating the MicroPlate with the cell suspension, they have most likely been heat damaged.

Q. Do I have to handle frozen or lyophilized cultures in a special way?

A. Yes. Subculture frozen or lyophilized cultures two to three times before testing but no more than 5 times.

Q. Why is there a defined range for the turbidity of inocula?

A. It is essential to prepare inocula in a consistent and precise manner. The Biolog redox test chemistry is sensitive to oxygen concentration, which is determined by cell density. Use Biolog turbidity standards and check the calibration of the turbidimeter.

Q. Are Biolog's Turbidity standards comparable to McFarland standards?

A. Biolog's Turbidity standards have different set points and are not comparable to McFarland standards. Biolog's standards are set for cell densities at which the Biolog test performs optimally. Use of Biolog standards is necessary to achieve consistent and accurate results.

Q Are there any genera where the taxonomy is still undergoing change?

A. Taxonomists have not yet agreed upon how all species should be delineated and microbiology is an ever-evolving science. We update and expand our software library on a continual basis, but at this point there are certain genera still undergoing revision. These include Bacillus, Corynebacterium, Enterobacter, Pseudomonas, Aeromonas, Vibrio, Acinetobacter, Moraxella, Clostridium, and Candida among many others.

www.dsmz.de/bactnom is a good site for current bacterial nomenclature. You can also link to this site through Biolog's web page. www.cbs.knaw.nl is a good site for current yeast and filamentous fungi nomenclature and information.

Remember, Biolog Technical Services may be able to assist with any identification questions.

Q. Our lab is interested in your quality control and validation procedures. Where do we get this information?

A. Refer to the package insert and call Biolog Technical Services. Request our Validation Package information, Material Safety Data Sheets, Quality Control organism information, and Certificates of Performance.

Section 12. Troubleshooting

Carefully following the instructions in this guide will greatly minimize problems. Occasionally, however, you may get stuck or encounter difficulties. This section addresses the symptoms, causes, and solutions to those occasional problems. Call Biolog Technical Service if further assistance is required.

Culturing Microorganisms

Table 19 Culturing Microorganisms

Symptom	Cause	Solution
Poor overall growth	Using non-recommended media	Use Biolog-recommended media.
	Slow grower	Re-streak heavily (as a lawn) onto one or more agar plates. Incubate for 4-48 hours. This should give enough growth to inoculate the panel.
Isolate takes several days to form a colony	Some environmental organisms take several days to become visible on growth plate, at which point culture may be too old for successful ID	Subculture one or two passages in broth medium and centrifuge cells. Wash them prior to preparing inoculum. Set up two or three agar plates to obtain sufficient growth.
Bacterium will not grow on BUG + B or forms pinpoint colonies	Fastidious gram-negative bacteria need special culture conditions	Use Chocolate agar and incubate with elevated CO_2 at 35-37° C.
	Some environmental bacteria may be oligotrophic or temporarily in an oligotrophic state (i.e., they will only grow on low nutrient media such as R2A)	Try to subculture the bacterium from R2A and see if they will gradually adapt to growing on BUG + B. If not, it is a species that is not included in our Biolog database. Agricultural bacteria may be grown on BUG without blood.
Slow growth and/or weak pattern formation	Sub-optimal growth temperature and/or humidity and/or atmosphere	Use specified incubating temperatures (see Section 4 and Appendices). If the unknown organism came from a cold or warm environment, grow it first at its environmental temperature, then try to grow it at 35-37° C, 30° C, or 26° C. Add a pan of water to provide humidity in your incubator. Incubate the MicroPlate at the same temperature as the growth plate.

Table 19 Culturing Microorganisms

Symptom	Cause	Solution
Mixed growth on agar plates	Sticky bacterial surfaces	Transfer a colony into a few ml sterile water, vortex for several seconds, and streak out from the cell suspension onto a medium that aids the detection of subtle differences in colony morphology. Take a single colony and streak for isolation on a separate plate. Repeat as necessary until you have a pure culture.

Preparing Inocula

Table 20 Preparing Inocula

Symptom	Cause	Solution
Incorrect turbidity	Turbidimeter out of calibration	Check calibration accurately. Use correct standards. Make sure standards have not expired. *For annual preventive maintenance contracts contact Technical Services.
	Tubes not properly blanked	Once you blank a tube, do not rotate it while making suspension. Blank each suspension tube (tubes are not optically precise and can vary tube-to-tube and with rotation)
	Significant scratches or smudges on tube	Use suspension tubes only once, then discard. Visually inspect tubes before using. Wipe outside of tubes with a tissue before placing in turbidimeter.
Inaccurate %T measurements	Nonuniform suspension of cells	Use Inoculatorz to mix cells all the way to the bottom of the tube. The light path that looks through the tube is only viewing the bottom of the tube.
Trouble making a homogenous suspension	Mucoid or clumpy cells	1. Roll swab over young colonies in 3rd and 4th quadrants. 2. Mash colonies against side of a sterile tube (above meniscus). 3. Add several ml of inoculating fluid and wash the sides of the tube with a cotton swab. 4. Mix well, then examine for clumps. If clumps are present, allow them to settle. 5. Transfer this concentrated inoculum into a fresh Inoculating Fluid tube until the recommended turbidity is reached. * Be careful not to transfer any colony clumps into the Inoculating fluid.

Table 20 Preparing Inocula

Symptom	Cause	Solution
		1. For difficult organisms such as *Gordonia* or *Tsukamurella*, suspend colonies as a dense suspension in a small amount of inoculating fluid as above. 2. Incubate as needed at 35° C (do not exceed 10 minutes). Vortex (if clumps still present, pull off supernatant and repeat). 3. Bring supernatant volume up to 18 ml and vortex until homogenous.
		Use suspension tubes only once, then discard.

Inoculating MicroPlates

Table 21 Inoculating MicroPlates

Symptom	Cause	Solution
Pipetting problems	Wells inoculated unevenly	Dispense first aliquot from pipettor back into reservoir. Depress lever smoothly and completely. Make sure tips are lined up evenly and tightly seated. Avoid pipetting or trapping air bubbles. Visually check wells after inoculating. *For annual preventive maintenance contracts contact Technical Services.
	Incorrect volumes used	100 µl for aerobic bacteria.

Incubating MicroPlates

Table 22 Incubating MicroPlates

Symptom	Cause	Solution
Poor growth or poor pattern formation in MicroPlates	Wrong incubation conditions Wrong MicroPlate	Incubate according to specified temperature and conditions. See Section 5 and Appendices. Incubate the MicroPlate at the same temperature as the growth plate. Make sure incubator humidity is sufficient.

If all wells in columns 1-9 are positive	Bacterium is a strongly reducing species causing false positive color in the A-1 well	Repeat the test using Protocol B and IF-B.
	Inoculum density is not excessive or too clumpy	Check the calibration of the turbidimeter. *For annual preventive maintenance contracts contact Technical Servies.
	A-1 well is not under-filled	It is used as a reference well. Retest.
If all wells in columns 1-9 are negative	Oligotrophic species or extremely slow growing bacteria, for example, may give all negative wells	Incubate off-line longer and perform immediate read. Organism may not be identifiable on the system.
	Cells are not freshly grown	Restreak and retest.
	A-1 well is under or over - filled	A-1 is used as a reference well. Retest.
	Incubation temp and atmospheres not correct	Restreak under the correct conditions and retest.
	Inoculum density is too low	Check the calibration of the turbidimeter.
	Inoculating fluid conditions not appropriate	Check that inoculating fluid was stored correctly and was prewarmed prior to use.

OmniLog Incubator/Reader

Table 23 OmniLog incubator/Reader

Symptom	Cause	Solution
Erratic or inaccurate reading	Moisture, scratches, or smudges on MicroPlate	If using an off-line incubator, wipe bottom of MicroPlate before putting into incubator/reader.
	Many borderline reactions on MicroPlate	Review all sample preparation procedures for correctness and accuracy.
Software won't communicate with or initialize incubator/reader	Wrong com port selected	Choose correct com port.
	Loose cable connection	Turn incubator/reader off. Unplug cable, then plug it back in. Turn incubator/reader on and try again.
	Unknown cause	The incubator/reader has its own error messages, which should be self-explanatory. Call Biolog Technical Service if you need further assistance.
MicroPlate won't go into incubator/reader	MicroPlate mispositioned	Make sure MicroPlate into place and is seated levelly. Make sure MicroPlate lid is on. Remove MicroPlate and reposition in incubator/reader tray. Make sure A1 well is at the right rear. Verify that software is communicating with incubator/reader.
	Camera assembly is latched onto that row	Click Move Camera selection bar on the Load Plates window to move camera to a different row.
Software can not contact the incubator/reader via the serial port	Loose cable or incorrectly installed cable	Make sure the serial cable is connected to serial port No. 1 on the incubator/reader (this will be the lower plug).
No video signal (footer bar error message "Video Error")	Loose or incorrectly installed cable	Click on the "Video Error" message on the footer bar. If you get a message reading "No Signal", check the video cable at the back of the incubator/reader. The cable should go from a special plug at the back of the computer to the upper plug at the back of the incubator/reader.
Specified error message in footer bar	Hardware Jam, Power or Video failure.	Write down the message and call Biolog Technical Services.
"Not at temperature" error message in footer bar and "Not at Temp" light in front of instrument goes on	Set and actual temperature out of range (+/- 2°C)	If there is more than a 2°C difference between the temperature you set and the actual temperature of the incubator/reader, this message will appear. This message will appear whenever you re-set the temperature and remain in place until the temperature range has been reached. The "Not at Temp" light at the front of the instrument will also illuminate until the temperature range has been reached.

Table 23 OmniLog incubator/Reader

Symptom	Cause	Solution
"Interrupt On" light at front of incubator / reader goes on.	You have tried to open the door while the camera assembly is moving.	Shut the door immediately. The incubator/reader will reset. To avoid making this error again, keep an eye on the footer bar for the message "Reader OK, Busy". When this message shows, do NOT open the door.

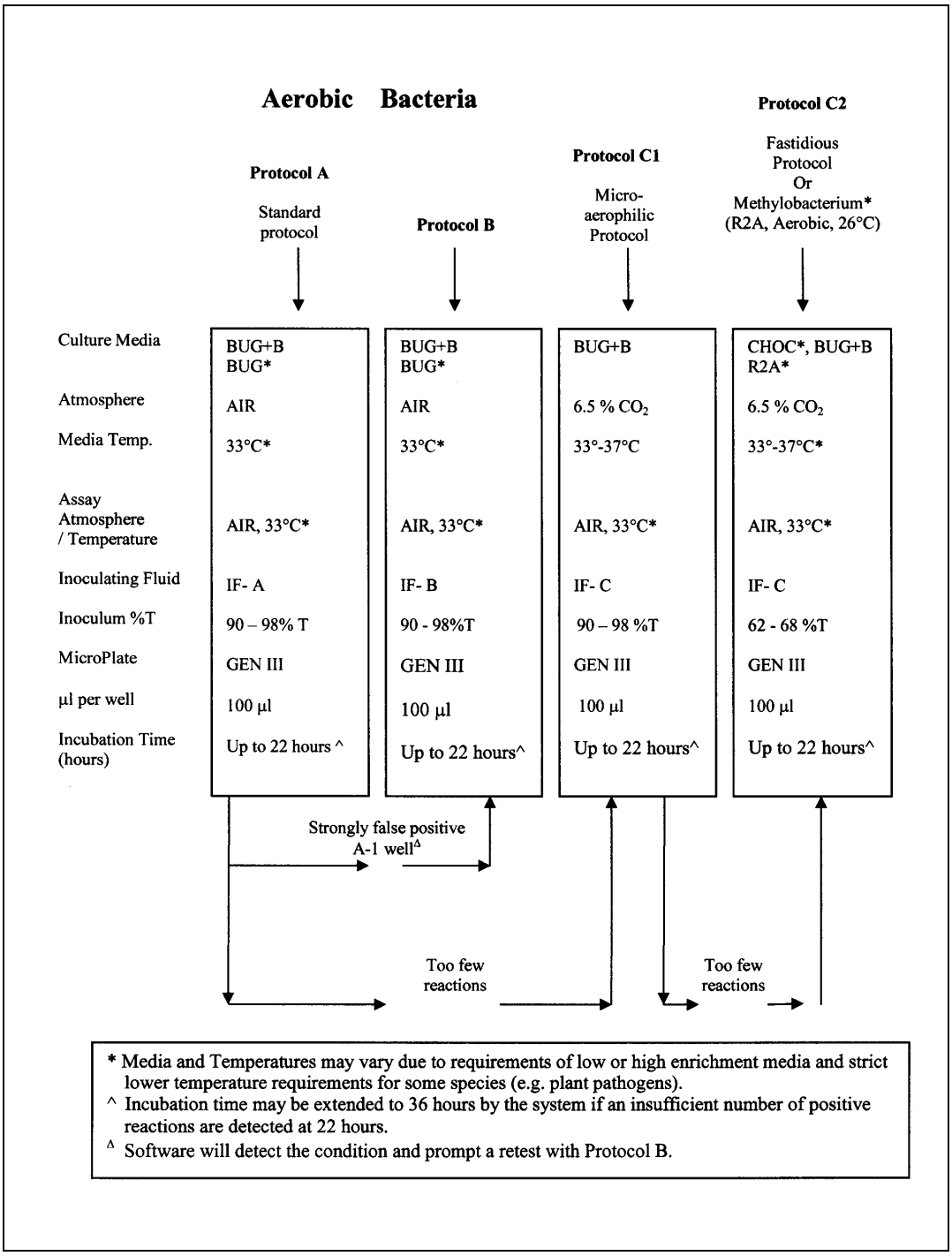
Section 13. Glossary

Aseptic technique	Standard lab procedures used to prevent contamination.
Carbon source utilization	Basic process used to identify microbes based on the chemicals they can utilize.
Dendrogram	Cluster diagram analysis in the form of a branching tree.
Enteric	Gram-negative bacteria belonging to the group Enterobacteriaceae.
False Positive Call	If the Raw A1 value is >= 125 and the current number of positives is more than 5 below the maximum number of positives for the plate so far, or if the Raw A1 value is >= 225.
Freeze	The act of converting a data file into a Frozen and non-editable format.
Genus ID	If the plate is on the final read (22 or 36 hours) and the first choice SIM does not exceed 0.5, but the sum of the top choice species SIM values exceeds 0.5 and are in the same genus.
Gram negative	Bacteria showing typical pink or red reaction on Gram stains.
Gram positive	Bacteria show typical blue or violet reaction on Gram stains.
Histogram	Visual representation of MicroPlate color distribution and thresholds.
Inocula	Cell suspension used to inoculate MicroPlates.
Inoculating fluid	Fluid used to prepare inocula.
Insufficient Pattern	If the plate has either (a) fewer than 2 positive metabolic reactions, or (b) the first choice ID does not match the previous read first choice ID, or (c) the first choice SIM is less than 0.2.
GEN III MicroPlate	Plate with 94 phenotypic tests: 71 carbon source utilization tests, with A1 well as the negative control and 23 chemical sensitivity tests, with A10 as the positive control.
MicroPlate reactions	Positive, negative, and borderline color response interpretation used to identify microbes.
Non-enteric	Gram-negative bacteria not belonging to the group Enterobacteriaceae.
Pattern	Color responses in MicroPlates.
Pleomorphic	Having various distinct forms or shapes exhibited by a single strain or species

Progressive ID (PID)	Biolog, Inc. developed pattern matching method which considers the progressive sequence in which purple wells are formed.
Pure culture	Culture containing only one microbial species.
Restricted Access Mode	Mode set by system administrator to prevent unregistered users from using system; assign user names, passwords and privileges to each user; create an audit trail, and freeze data files to maintain data integrity.
Species ID	If the plate has a first choice SIM that exceeds that threshold in two consecutive reads.
Sufficient Pattern	If the plate has (a) 2 or more metabolic positive reactions, and (b) the first choice species has matched a previous read, and c) a first choice SIM is greater than or equal to 0.2 but below the threshold. (Minimum requirement for an ID call attempt or for building a User database.)
Thresholds	The optical boundaries between negative, borderline, and positive reactions.
Too Many Borderline Reactions:	if there are >25 borderline reactions on the plate.
Turbidity	Measurement of cloudiness, which is indicative of inocula cell densities.
Unrestricted Access mode	Mode set by system administrator to allow any user to use all OmniLog software functions.
User Database	Database produced by the User, developed by using edited User data files to designate the reaction patterns of organism group clusters for specialized identification purposes. (Use RetroSpect Trending and Tracking Software)

Section 14. Appendices

Appendix 1: GEN III Protocol Flowchart



* Media and Temperatures may vary due to requirements of low or high enrichment media and strict lower temperature requirements for some species (e.g. plant pathogens).
^ Incubation time may be extended to 36 hours by the system if an insufficient number of positive reactions are detected at 22 hours.
Δ Software will detect the condition and prompt a retest with Protocol B.

Appendix 2: OmniLog GEN III Sample Batch sheet

Tray No.	Protocol	Sample ID				
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						
A						
B						

Appendix 3: Software ID Printout

Reader printout (Basic ID result sample)

1➔	ProgramName	OmniLog 2.1.01.136
	ProjectName	OL3
2➔	DataFileName	OL3_196_080418_N.D5E
	DataPath	C:\Documents and Settings\Administrator.TAS-3749COHWF1F\Desktop
3➔	WSOperator	J Smith
	Plate Errors	
	DataMode	ID
4➔	ReadMode	Single Read ID
5➔	Setup Time	Apr 17 2008 2:57 PM
6➔	Maximum Incubation Hours	22
	Current Incubation Hours	22.00
7➔	Reader	196
8➔	Position	20B
9➔	PlateType	GEN III
	Protocol	A
10➔	Sample ID	648993E
	Sample Source	BUILDING 45
	Notes	RETEST
	MicroPlate Lot No.	2387649
	Inoculation Fluid Lot No.	098747623
11➔	ID Date Time	Apr 18 2008 1:00 PM
12➔	Biolog ID DB	C:\Program Files\Biolog\OL_DB_dir\databases\Biolog GEN III DB.I5G
	ID State	Final ID
	ID Result	Species ID: Escherichia coli
13➔	ID Organism Type	GN-Ent
	ID Comment	
	ID Notice	
16➔	Report Date	Apr 18 2008 1:01 PM

Reader printout (ID result with optional view ID list sample)

1➔	ProgramName: OmniLog 2.1.01.136
2➔	ProjectName: OL3
	DataFileName: OL3_196_080418_N.D5E
3➔	DataPath: C:\Documents and Settings\Administrator.TAS-3749COHWF1F\Desktop
	WSOperator: J Smith
	Plate Errors
	DataMode: ID
4➔	ReadMode: Single Read ID
5➔	Setup Time: Apr 17 2008 2:57 PM
6➔	Maximum Incubation Hours: 22
	Current Incubation Hours: 22.00
7➔	Reader: 196
8➔	Position: 20B
9➔	PlateType: GEN III
	Protocol: A
	Sample ID: 648993E
10➔	Sample Source: BUILDING 45
	Notes: RETEST
	MicroPlate Lot No.: 2387649
	Inoculation Fluid Lot No.: 098747623
11➔	ID Date Time: Apr 18 2008 1:00 PM
12➔	Biolog ID DB: C:\Program Files\Biolog\OL_DB_dir\databases\Biolog GEN III DB.I5G
	ID State: Final ID
13➔	ID Result: Species ID: Escherichia coli
	ID Organism Type: GN-Ent
	ID Comment
	ID Notice

Rank	PROB	SIM	DIST	Gram	Organism Type	Species
1	1.000	0.841	2.280	GN	GN-Ent	Escherichia coli
14➔ 2	0.000	0.000	7.216	GN	GN-Ent	Escherichia coli inactive
3	0.000	0.000	9.123	GN	GN-Ent	Escherichia coli O157:H7
4	0.000	0.000	9.894	GN	GN-Ent	Escherichia fergusonii

16➔ Report Date: Apr 18 2008 1:10 PM

Reader printout (ID result with optional view ID list sample and plate data value)

1➔	ProgramName: OmniLog 2.1.01.136
2➔	ProjectName: OL3
	DataFileName: OL3_196_080418_N.D5E
	DataPath: C:\Documents and Settings\Administrator.TAS-3749COHWF1F\Desktop
3➔	WSOperator: J Smith
	Plate Errors:
	DataMode: ID
4➔	ReadMode: Single Read ID
5➔	Setup Time: Apr 17 2008 2:57 PM
6➔	Maximum Incubation Hours: 22
	Current Incubation Hours: 22.00
7➔	Reader: 196
8➔	Position: 20B
9➔	PlateType: GEN III
	Protocol: A
10➔	Sample ID: 648993E
	Sample Source: BUILDING 45
	Notes: RETEST
	MicroPlate Lot No.: 2387649
	Inoculation Fluid Lot No.: 098747623
11➔	ID Date Time: Apr 18 2008 1:00 PM
12➔	Biolog ID DB: C:\Program Files\Biolog\OL_DB_dir\databases\Biolog GEN III DB.I5G
	ID State: Final ID
13➔	ID Result: Species ID: Escherichia coli
	ID Organism Type: GN-Ent
	ID Comment:
	ID Notice:

Rank	PROB	SIM	DIST	Gram	Organism Type	Species
1	1.000	0.841	2.280	GN	GN-Ent	Escherichia coli
2	0.000	0.000	7.216	GN	GN-Ent	Escherichia coli inactive
3	0.000	0.000	9.123	GN	GN-Ent	Escherichia coli O157:H7
4	0.000	0.000	9.894	GN	GN-Ent	Escherichia fergusonii

14➔

Data	1	2	3	4	5	6	7	8	9	10	11	12
A	133	{ 227 }	{ 217 }	< 237 >	144	{ 190 }	{ 192 }	{ 201 }	{ 183 }	< 303 >	< 304 >	< 307 >
B	{ 171 }	{ 182 }	{ 225 }	< 244 >	{ 164 }	< 241 >	< 255 >	< 248 >	< 254 >	< 291 >	< 276 >	{ 228 }
C	{ 214 }	{ 224 }	{ 212 }	< 228 >	88	{ 216 -	< 250 >	< 254 >	< 269 >	< 289 >	< 282 >	{ 249 -
D	< 255 >	{ 191 }	82	{ 161 }	< 268 >	< 280 >	< 273 >	{ 219 }	< 286 >	< 285 >	< 292 >	{ 188 }
E	104	< 256 >	< 281 >	{ 222 }	< 297 >	< 229 >	137	{ 183 }	< 268 >	< 297 >	< 283 >	< 299 >
F	{ 166 }	< 274 >	< 247 >	< 279 >	< 281 >	< 247 >	120	93	86	< 290 >	< 344 >	< 341 >
G	68	< 235 >	106	< 282 >	64	< 274 >	98	< 292 >	< 244 >	{ 219 -	{ 247 }	95 +
H	93	63	{ 225 }	{ 196 }	{ 209 }	107	< 281 >	< 290 >	{ 215 }	{ 258 }	< 300 >	< 265 -

15➔

16➔ Report Date: Apr 18 2008 1:04 PM

Report Printout Key

1. Software version
2. Data File Name
3. Operator
4. Read Mode
5. Time MicroPlate read was started *(Setup)*
6. ID read in hours
7. Omnilog Reader SN
8. MicroPlate Position
9. Plate Type and Protocol
10. Sample identifiers
11. Time of final read
12. Name of database(s) searched
13. ID result call box
14. Optional print: ID call list of 4 closest match species
15. Optional print: Numbers seen in each well is the Plate data Values. Threshold calls shown: Positive (<>), borderline ({}), and negative reactions (no brackets) for each type of assay (carbon source and chemical sensitivity)
16. Report print date and time.

Appendix 4: Footer Bar Entries

Cell	Message	Color/Action	Meaning
Door Status			
	Door Closed	Green	Incubator/reader door is closed
	Close Door	White	Close the door to maintain temperature and ensure future incubator/reader movement
Error Messages			
	No Errors	Green	No errors detected
	Hardware Abort	White	The incubator/reader is in mechanical failure
	Video Error	White	The camera failed during reading
	Temperature Error	White	The temperature is not at target setting while MicroPlates are in the incubator/reader
	Plate Error	White	A MicroPlate is missing from the read position
	Logged Auto Auto-Restart	White	Loss of computer power during read/incubation period. Number of restarts will appear in this box.
Reader Status			
	Reader OK, Idle	Green	The incubator/reader is not moving or reading, cycle on, reader idle.
	Reader OK, Busy	White/ Blinking	The incubator/reader is moving or reading, cycle on, reader busy.
	Air Read	White/ Blinking	The system is performing regular air read checks.
	Calibrating Lighting	White	The system is performing regular lighting calibration checks.
	Com Not Open	White	Serial port is not initialized
	Not Initialized	White	Incubator/reader is not initialized
	Cycle Off	White	The software is not in cycle mode
	Re-Trying Contact	White	The incubator/reader is not responding and the software is trying to re-establish contact
	User Cancel	White	User clicked “Stop Commands” / User time out commands

Cell	Message	Color/Action	Meaning
Temperature Status			
	Temp = XX.X	Green	This is the current incubator/reader temperature, which is in the target range
	Set T=33 vs T=XX	White	Temperature is out of range; shows target vs. actual temperatures
Plate Status			
	X Free, Y in Use	Green	Number of positions free and number in use
	X Plates Done	White	Number of MicroPlates ready to remove
Current System Time			
	Date and Time	Green	Date and time from operating system, updated every second (Month/ Day/ Year Hour: Minutes AM/PM)
User Name and Access			
	User: None	Green	No User is currently Logged in to the OmniLog Software
	User:A: "User Name"	Green	User is Logged in to the software. User name appears in this box along with a one letter code signifying that users access level. A: Administrator E: Edit User
	Unrestricted Access	White	Administrator has set software to Unrestricted Access mode. All Aspects of the software are available to all Users.
Time Until Auto Logout			
	Blank	Green	No User is currently logged into the software
		Grey Reset button Tan blinking countdown	15-1 countdown. Reset button resets countdown to 15 minutes. Auto Log-out reset also occurs when software is in use. When Tan box is blinking there are 10minutes or less to auto log-out.
Minutes Until Read			
	Blank	Green	No MicroPlates to read
	Min Until Read 12:30	Green	There are less than 15 but more than 10 minutes to the next read
	Min Until Read 9:30	Green/White blinking	There are less than 10 but more than 5 minutes to the next read
	Min to Read 2:30	White	There are less than 5 minutes to the next read

Cell	Message	Color/Action	Meaning
	Read In Progress	White	OmniLog is now reading
Time of Next Read			
	Blank	Green	There are no MicroPlates to read.
	Next Read X:XX AM/PM	Green	The next read.
	Snooze	Grey Button	The next read is in less than 5 minutes